

The multifaceted effects of Copper IUD on mucosal environment in African cisgender women

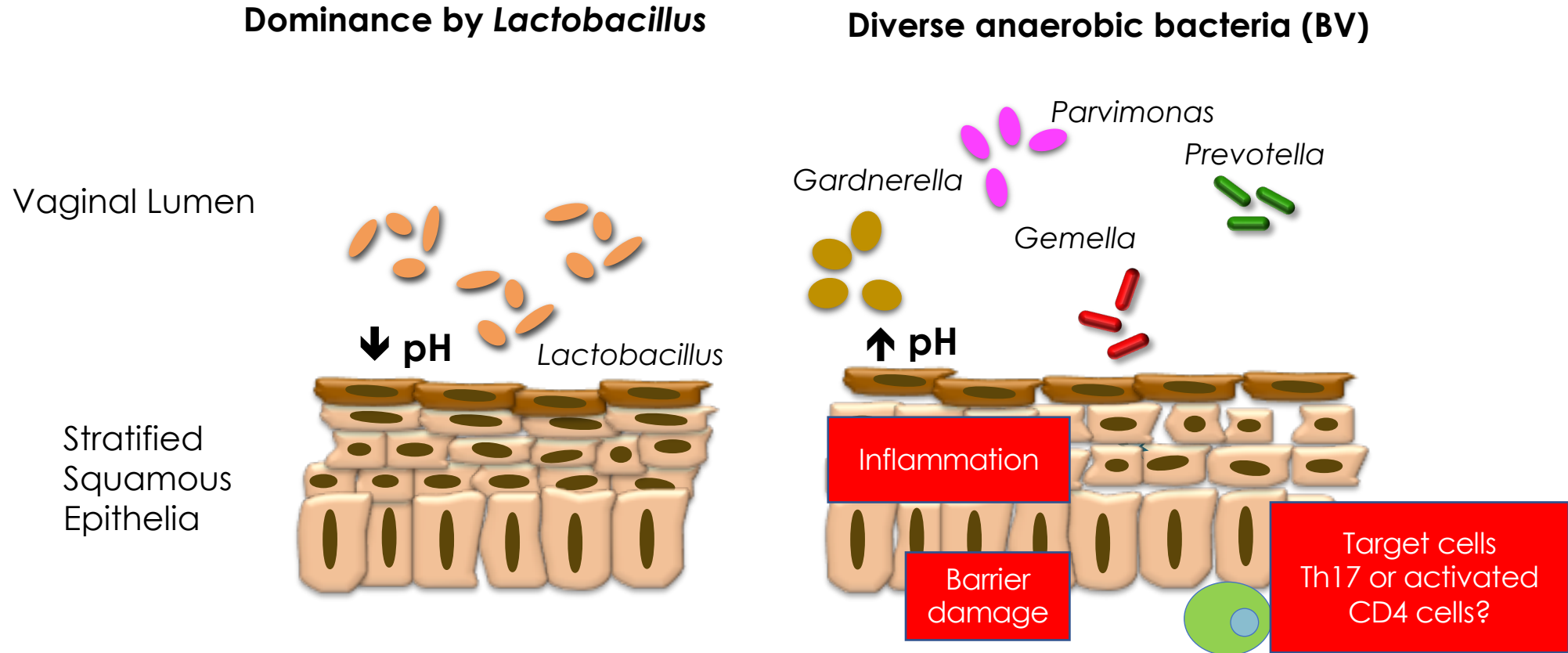
Heather Jaspán, MD, PhD



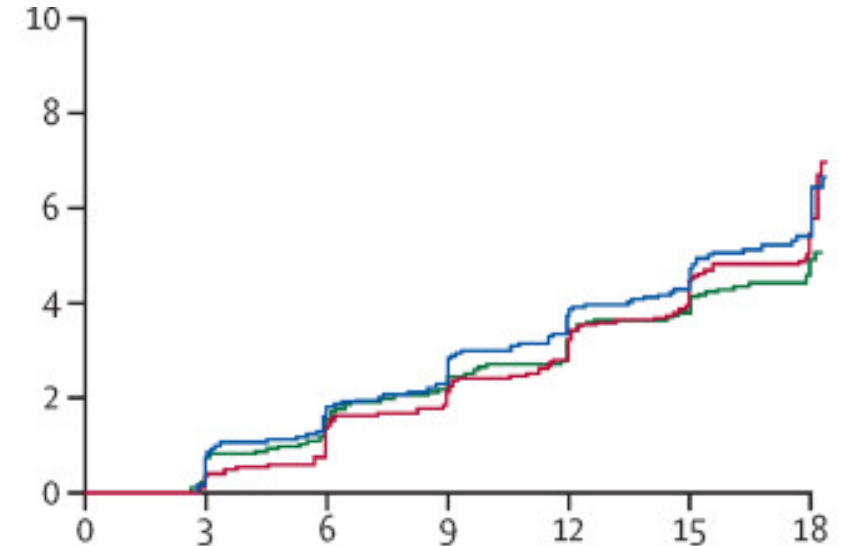
Contraception

- Contraception confers significant human rights and health benefits through reductions in unintended and high-risk pregnancies and maternal and infant morbidity and mortality
- Few contraceptive options available globally
- Women of reproductive age also at risk for HIV and STIs
- Observational studies suggested that hormonal contraceptives use may increase prevalence of HIV, STIs and bacterial vaginosis
 - Flawed by bias in condom use

High diversity communities (bacterial vaginosis) associated with increased risk of STIs and HIV



- ECHO: 7800 young adult women using contraception randomized to DMPA-IM or Copper-IUD
- Followed for up to 18 months to assess HIV incidence
- Only powered to detect 50% difference in HIV incidence
 - Found no significant difference in HIV incidence between arms;
 - Incidence was high (~4%)
 - Did not assess effect on other health outcomes such as BV and STIs

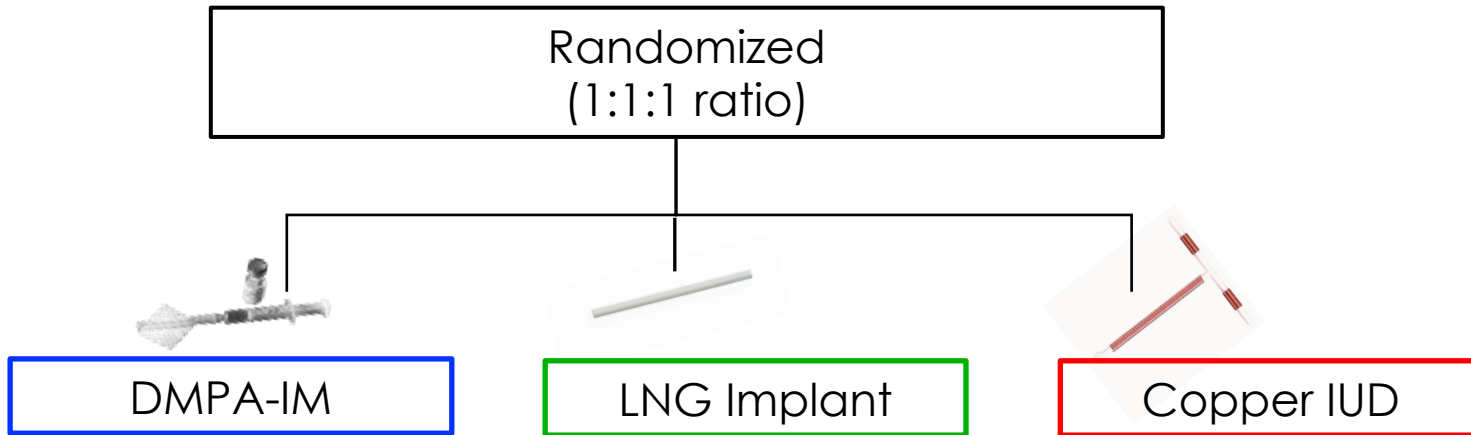


DMPA-IM vs copper IUD HR 1.04 (96% CI 0.82-1.33); p=0.72
 DMPA-IM vs LNG implant HR 1.23 (96% CI 0.95-1.59); p=0.097
 Copper IUD vs LNG implant HR 1.18 (96% CI 0.91-1.53); p=0.19

— DMPA-IM group
 — Copper IUD group
 — LNG implant group

Mucosal biomechanisms study nested within ECHO

Women co-enrolling in ECHO and ECHO BioMech studies at the Emavundleni (Cape Town), WRHI (JHB), and KEMRI (Kenya) sites



Vaginal swabs, cervical cytobrushes, soft cups at enrollment (pre-contraception) and 1 and 6-months (post contraception)

Stored specimens from each 3 monthly visit thereafter

Recalled ~ 2 years later: Vaginal swabs, cervical cytobrushes, pap smear: n=155

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(Heffron & Jaspan)

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(Heffron, Passmore & Jaspan)



Renee Heffron, PhD



Jo-Ann Passmore, PhD

Baseline cohort characteristics by arm

	Copper IUD (N = 62)	DMPA-IM (N = 64)	LNG Implant (N = 68)
Median age, years (IQR)	24 (21, 27)	24 (21, 26)	23 (21, 26)
Median BMI (IQR)	25.07 (21.56, 30.67)	24.52 (22.21, 30.30)	23.01 (20.96, 30.64)
Median gravidity (IQR)	1 (1, 2)	1 (1, 2)	1 (1, 2)
Median # primary partners (IQR)	1 (1, 1)	1 (1, 1)	1 (1, 1)
STI prevalence			
Chlamydia trachomatis, n (%)	14 (23)	9 (14)	5 (7)
Neisseria gonorrhoea, n (%)	5 (8)	3 (5)	3 (4)
HSV-2 seroprevalence, n (%)	25 (40)	34 (53)	30 (44)
Clinical BV prevalence, n (%)			
BV positive (Nugent 7-10)	20 (34)	15 (26)	21 (34)
BV intermediate (Nugent 4-6)	9 (15)	6 (10)	5 (8)
BV negative (Nugent 0-3)	30 (51)	37 (64)	35 (57)

High diversity, BV-associated vaginal microbiota were prevalent at baseline

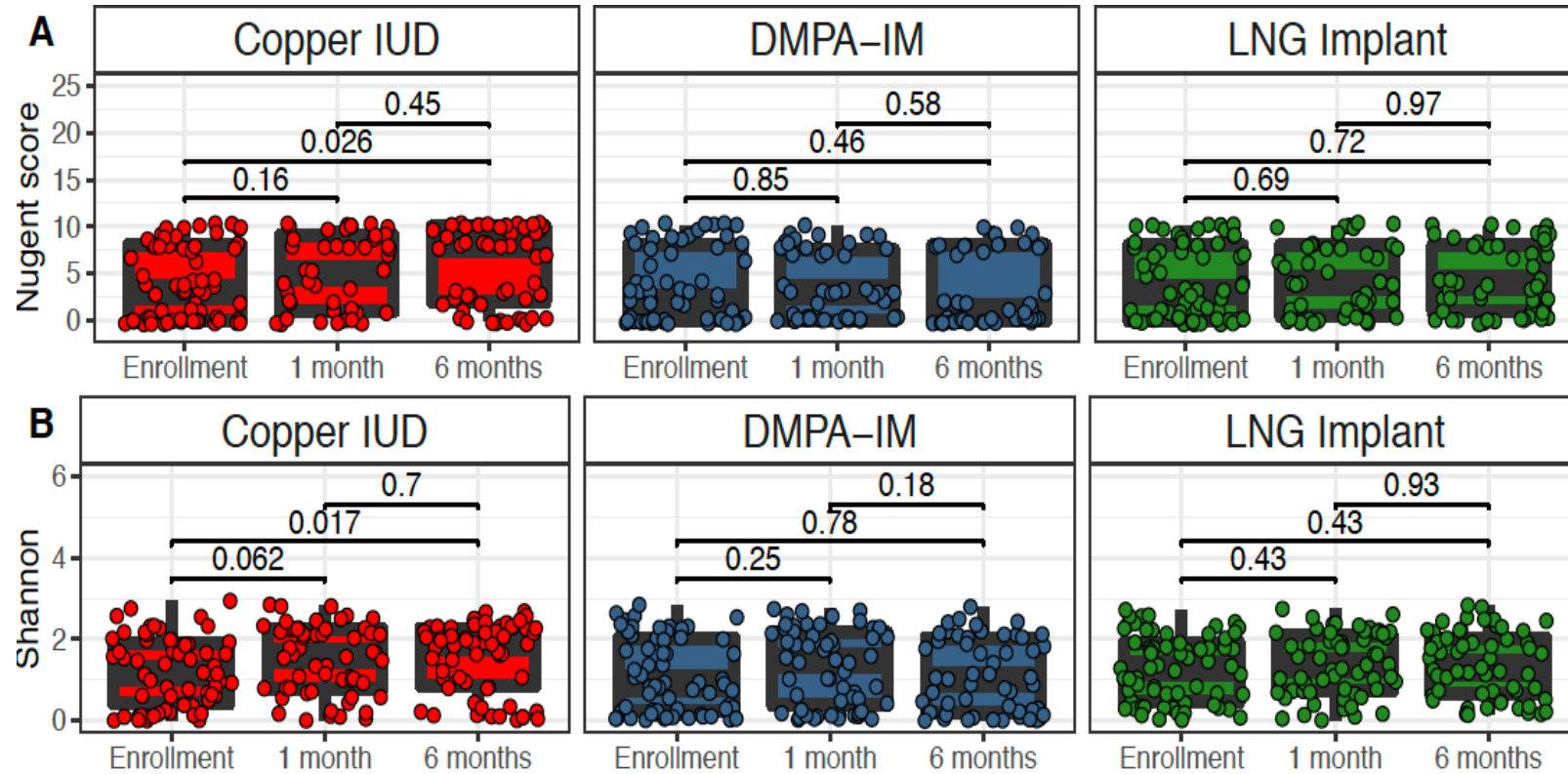


Nugent score: 0-3 No BV
4-6 Intermediate
7-10 BV

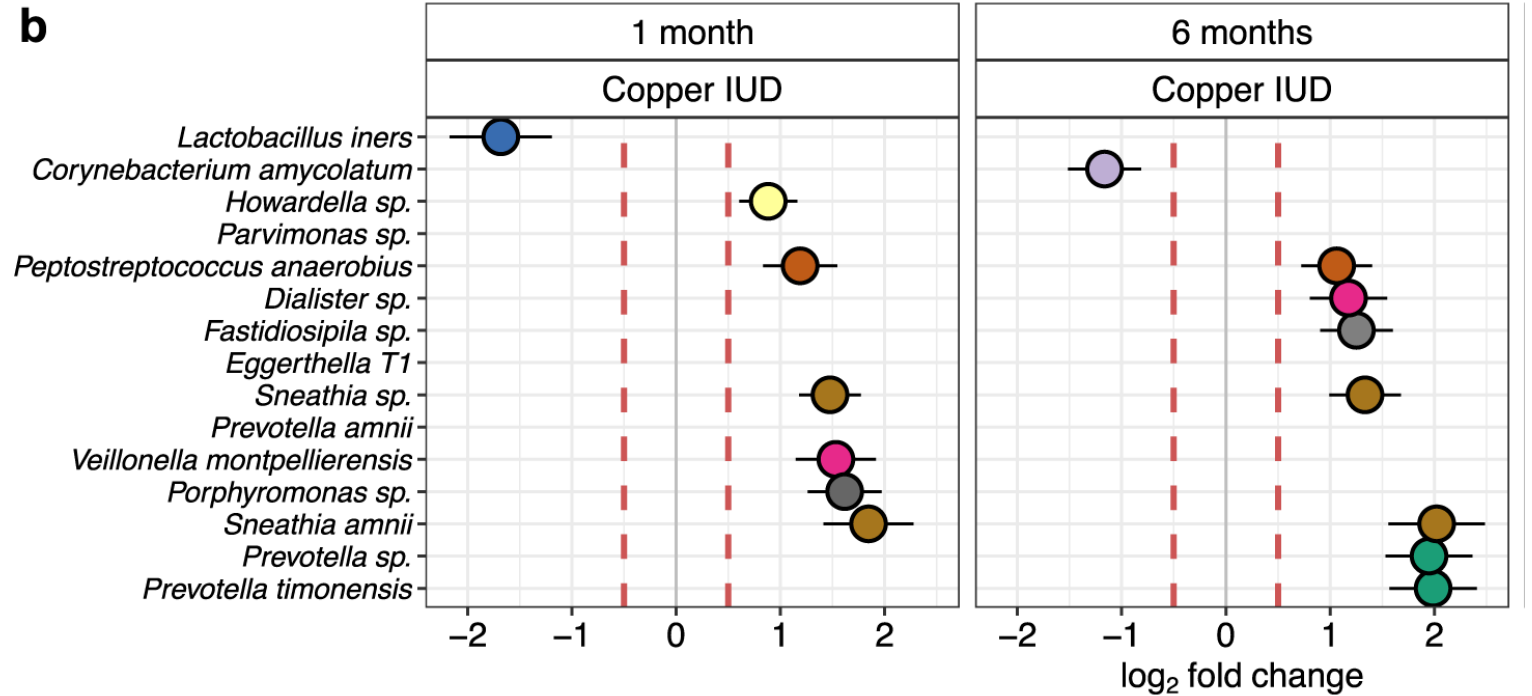
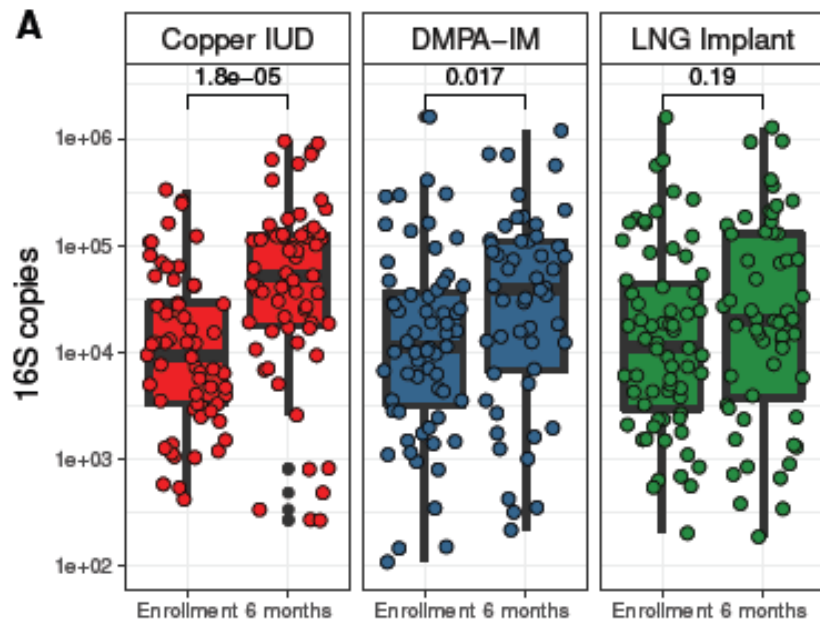
CST: I – *L. crispatus* dominated
III – *L. iners* dominated
IV - Diverse



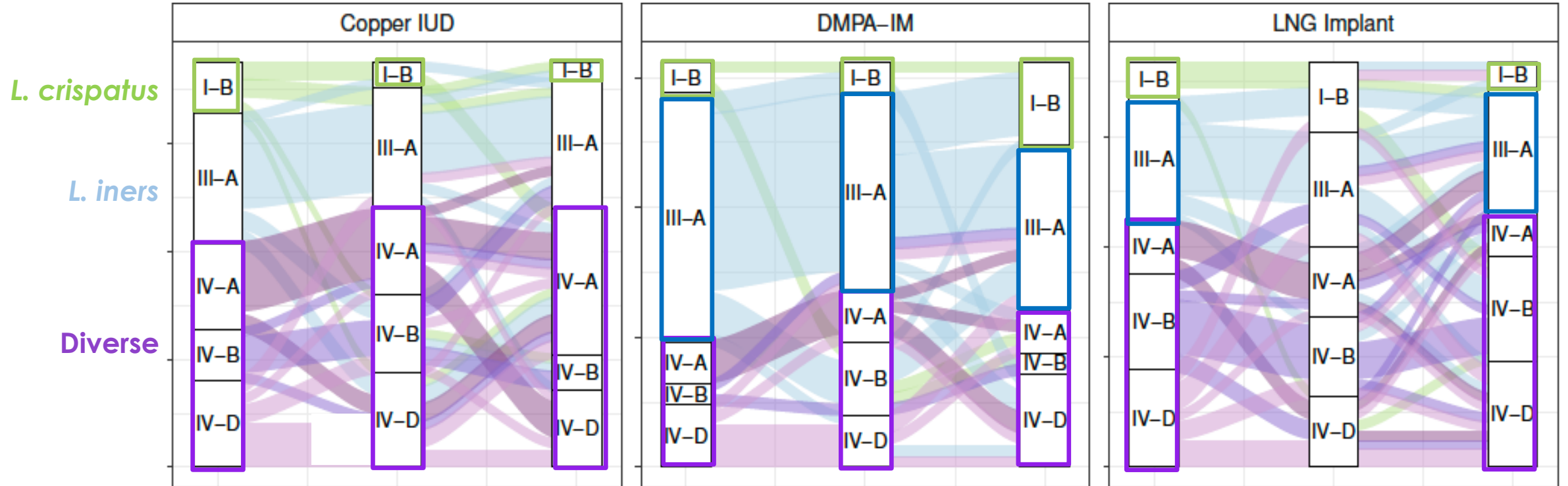
Copper-IUD increases vaginal microbiota diversity and clinical measures of BV



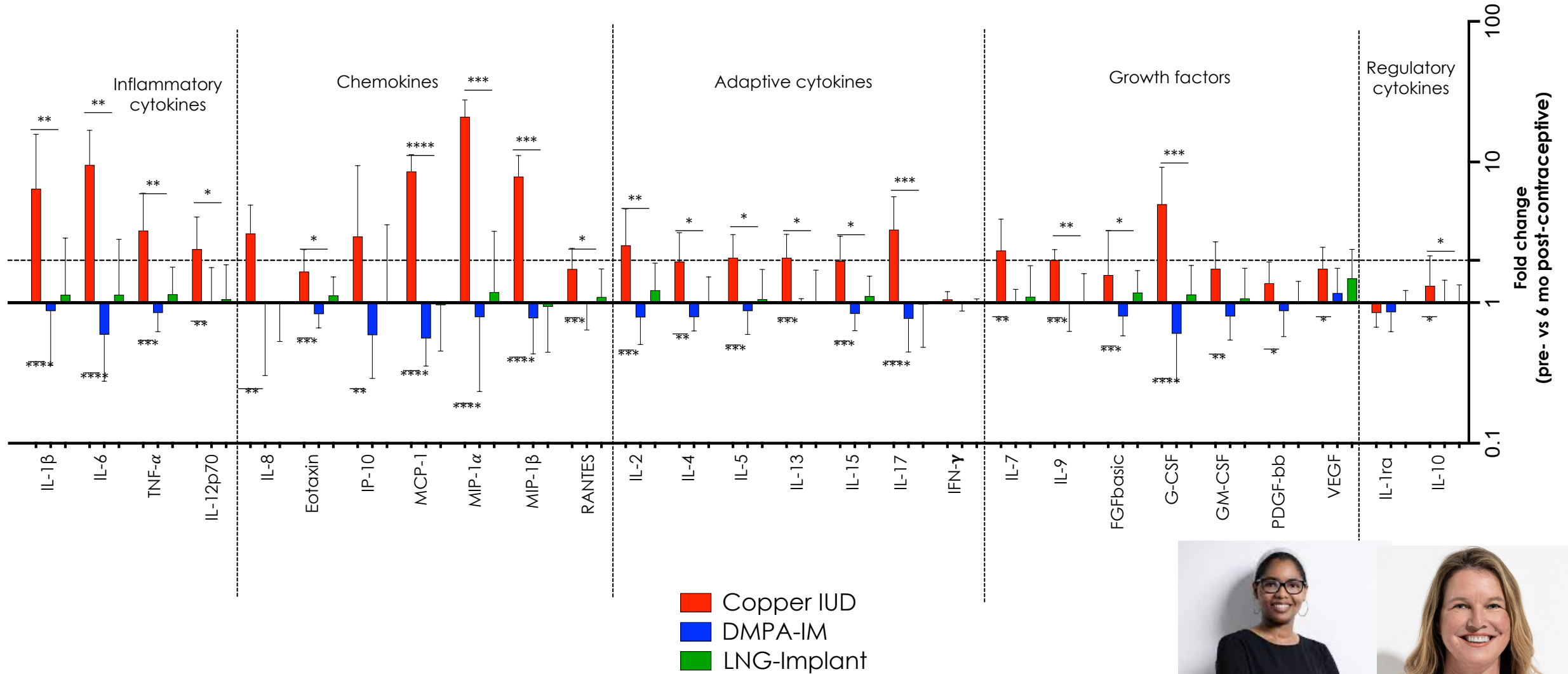
Cu-IUD increases bacterial load of facultative anaerobes



Cu-IUD induces shifts toward more diverse communities

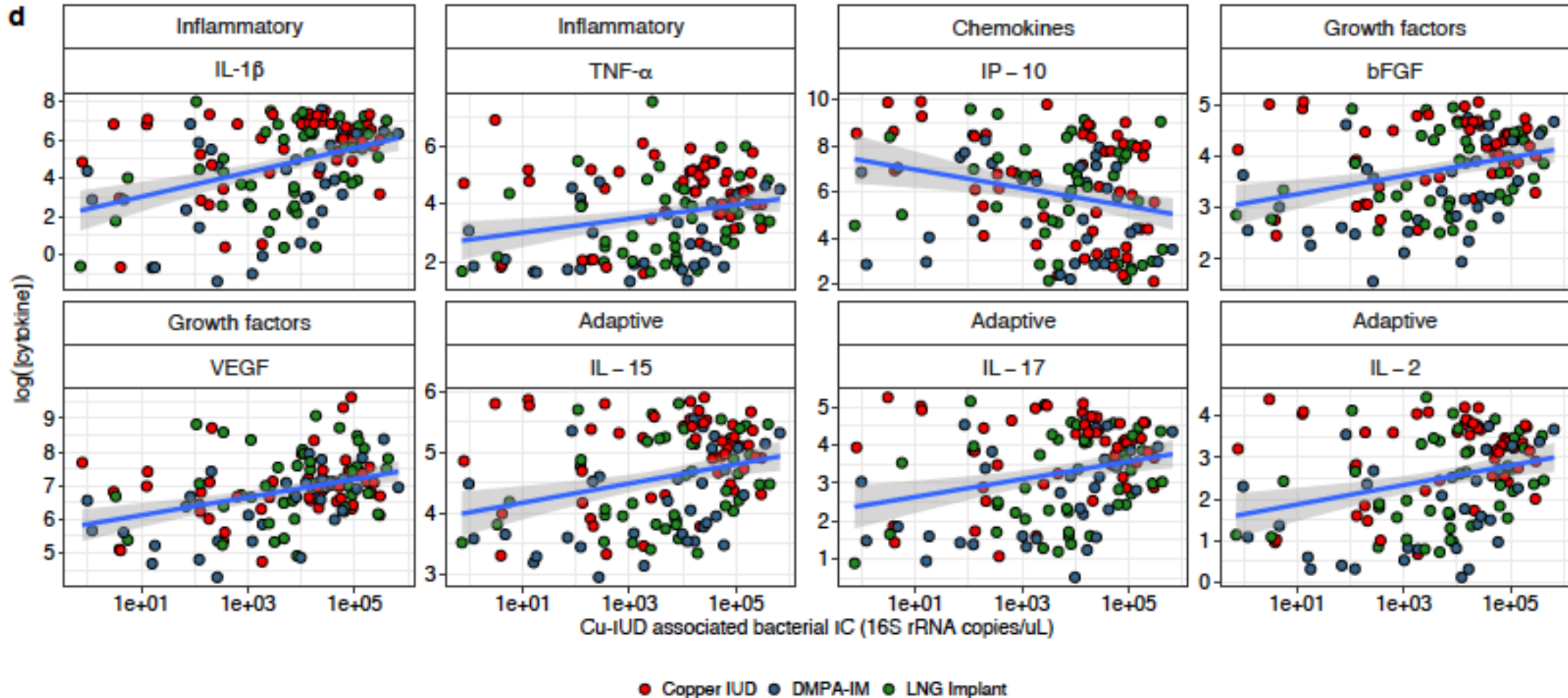


Copper IUD initiation induces genital inflammation

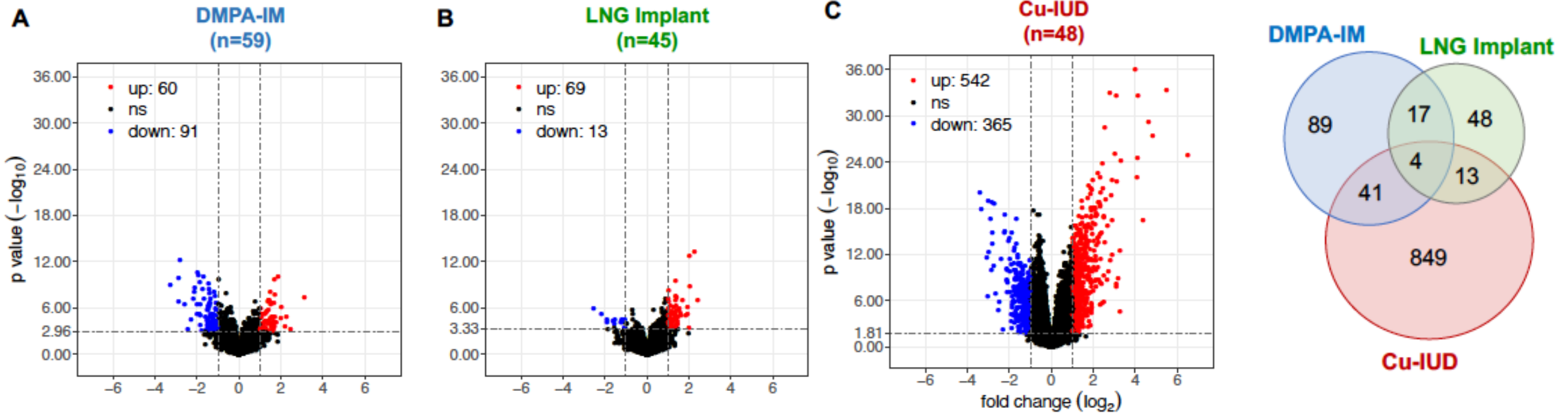


Tanko et al, *JID* 2022

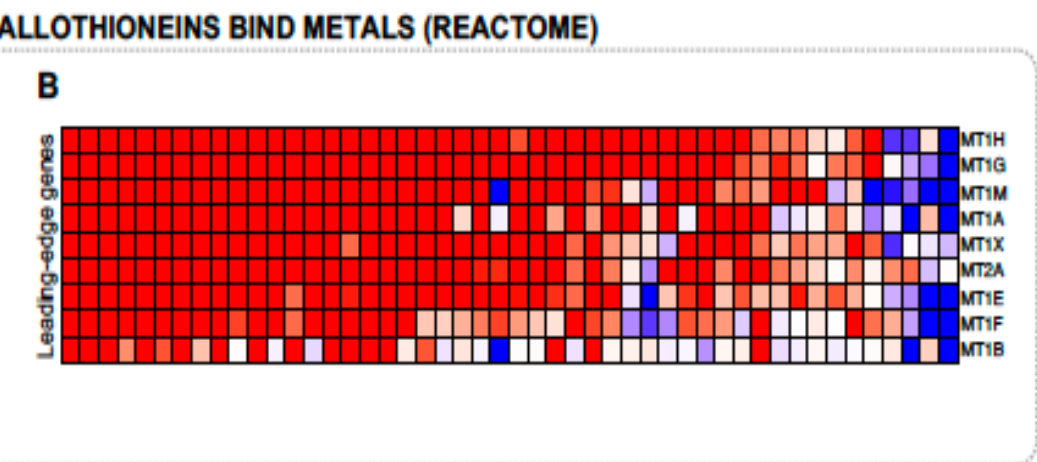
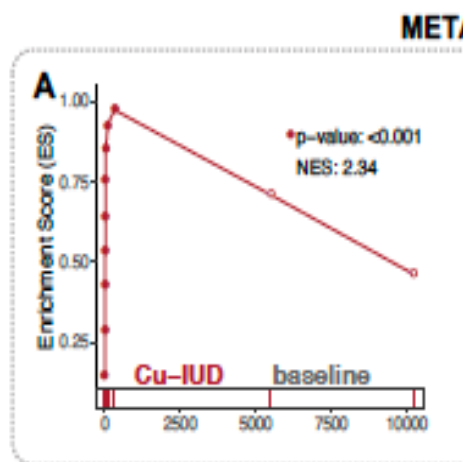
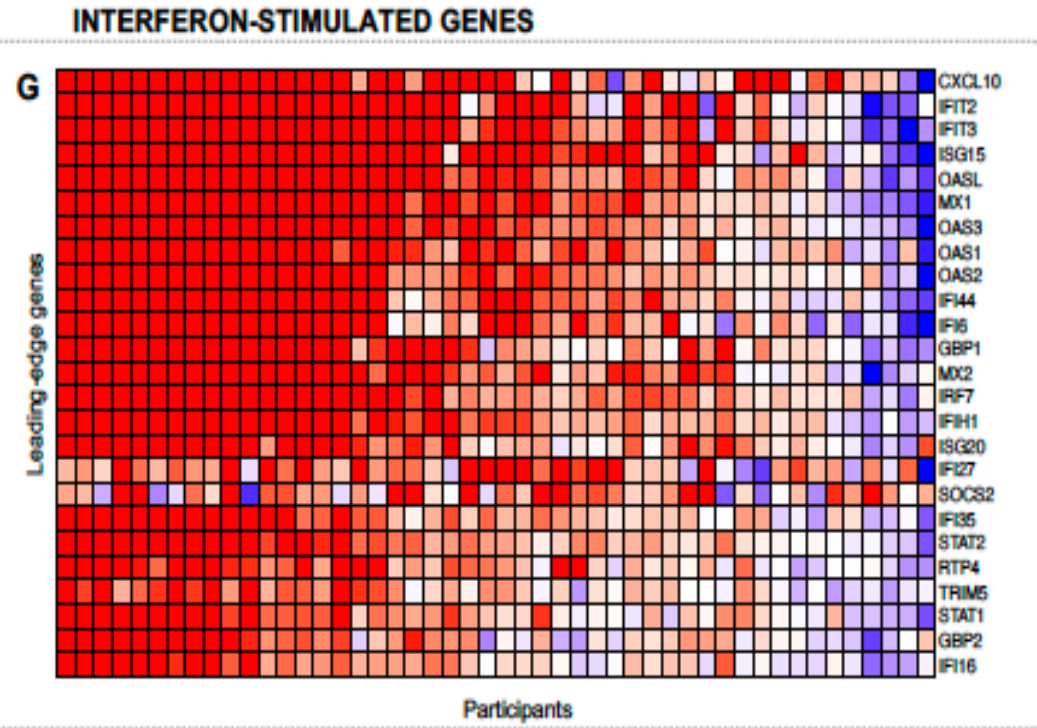
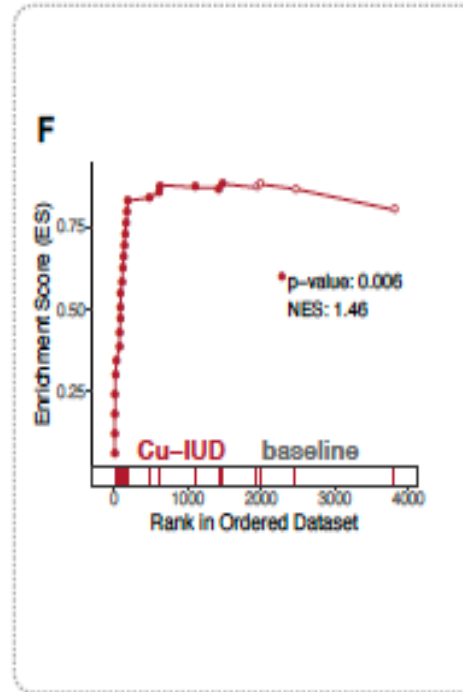
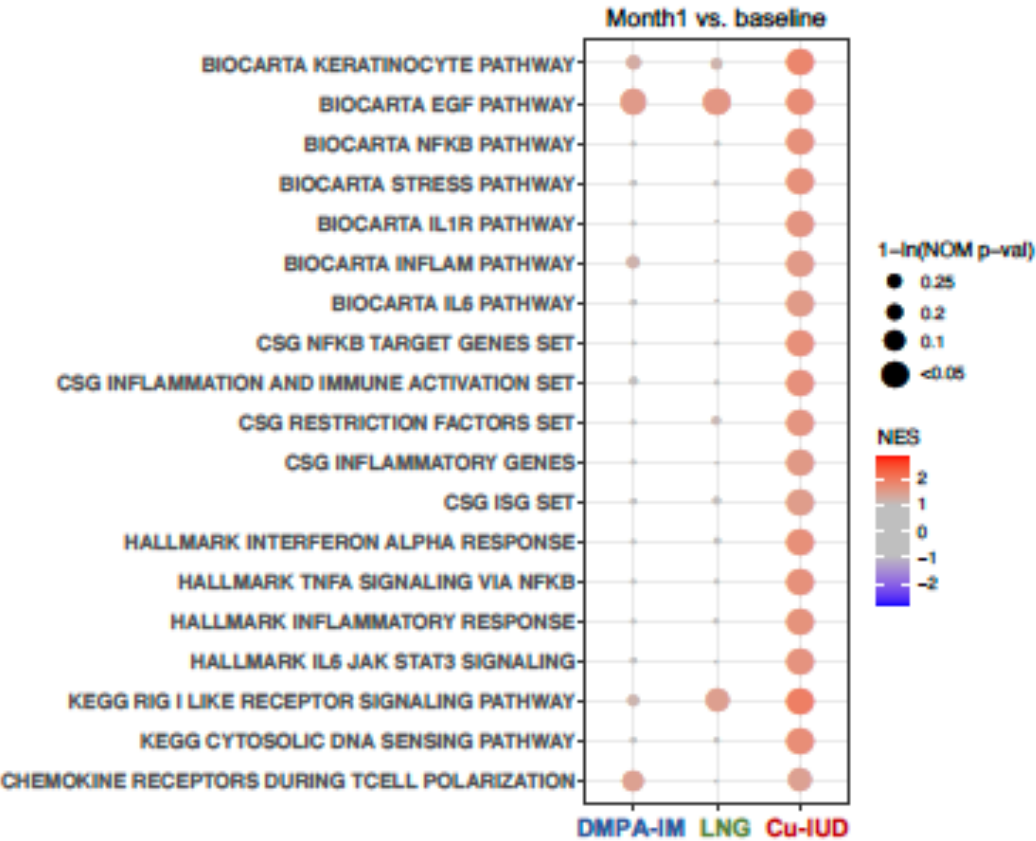
Genital inflammation strongly correlates with Cu-IUD bacterial load



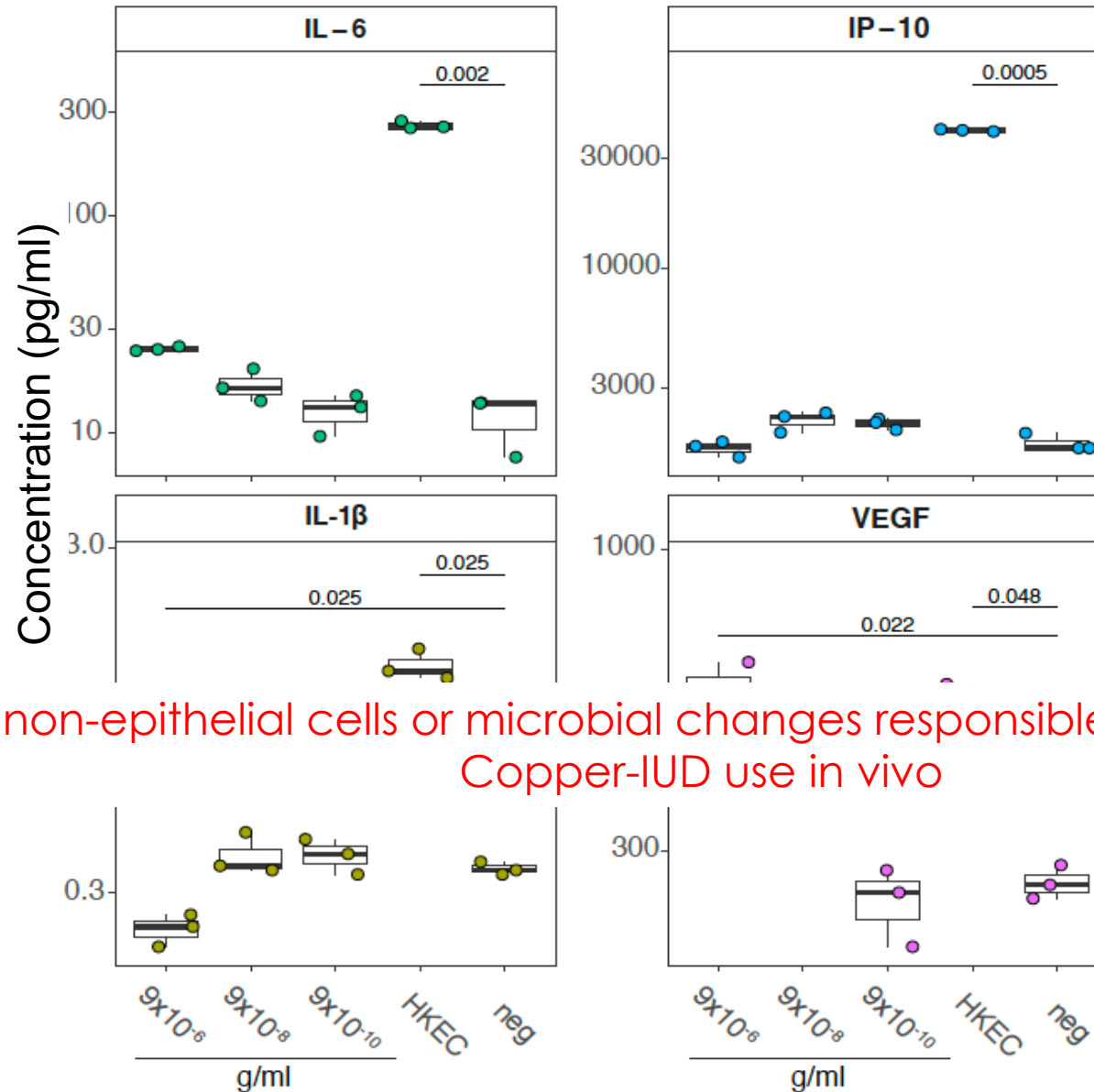
Copper-IUD induces large changes in cervical cell gene expression at 1 month post-initiation



Cu-IUD initiation induces inflammatory, stress and metallothioneine response genesets

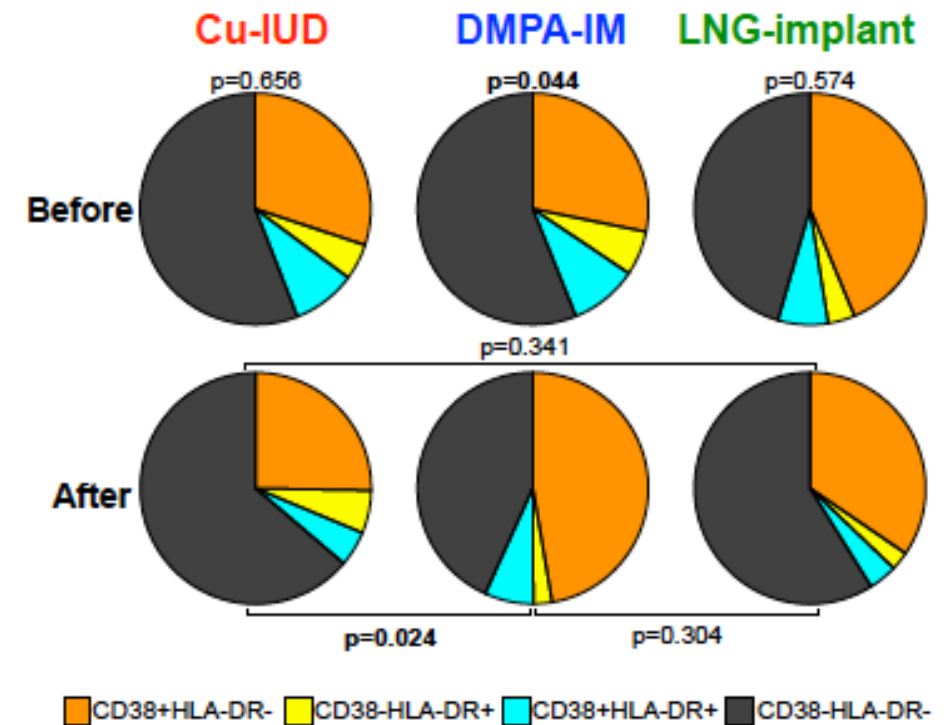
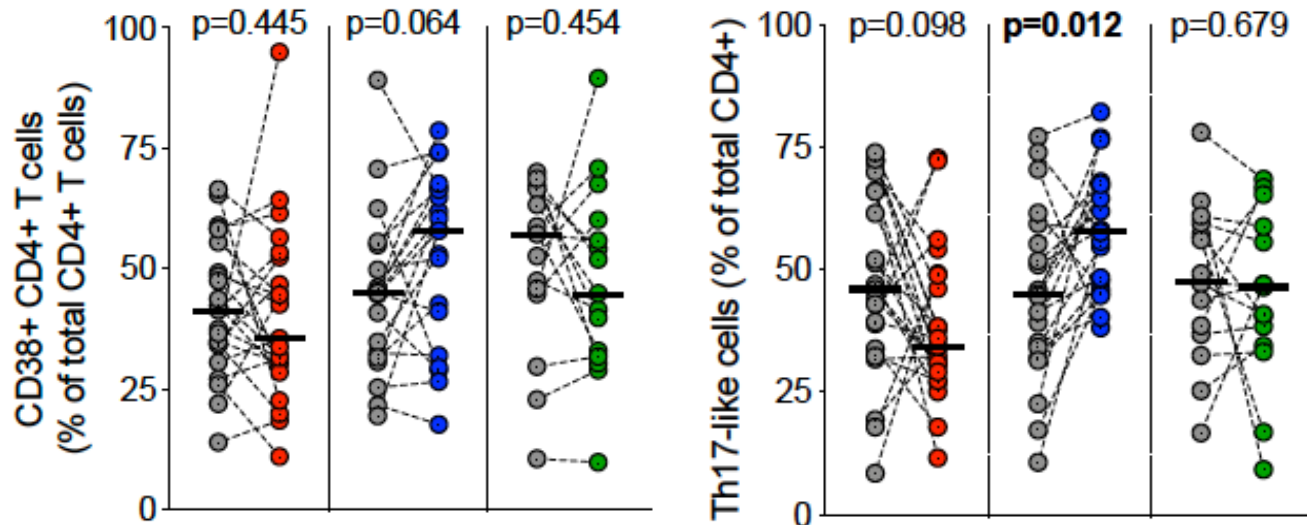


Copper induces VEGF but not inflammatory gene expression in endocervical cells



Possibly non-epithelial cells or microbial changes responsible for inflammation with Copper-IUD use in vivo

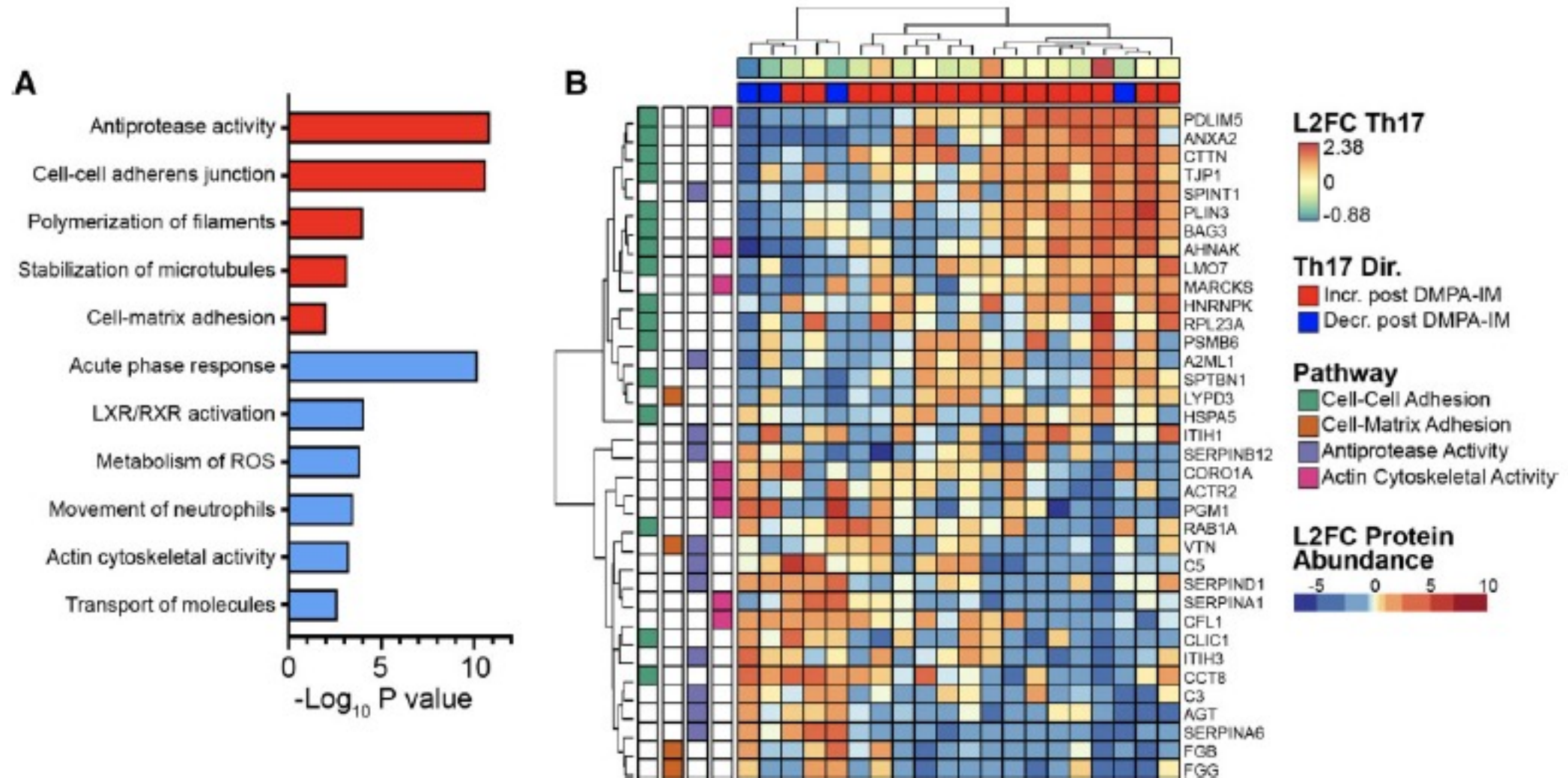
DMPA-IM induces HIV target cell activation in the cervix at 1 month post initiation



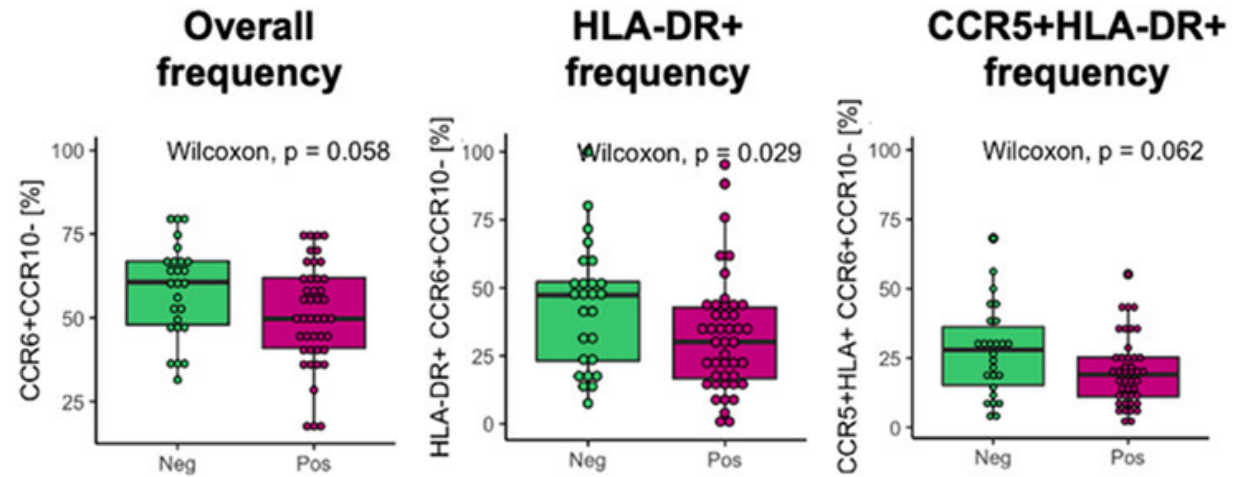
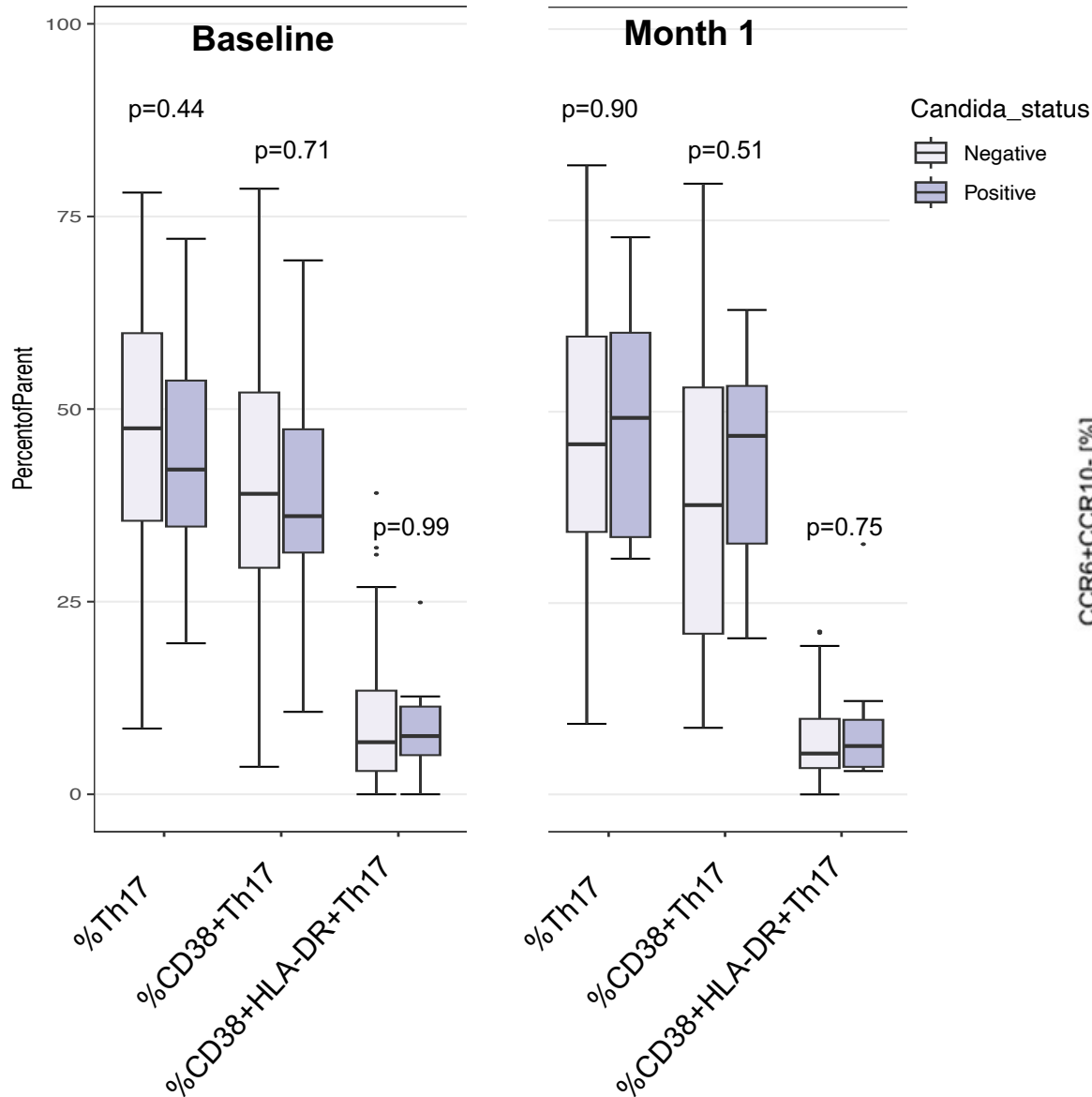
Th17 cells are important for mucosal barrier integrity and antifungal immunity



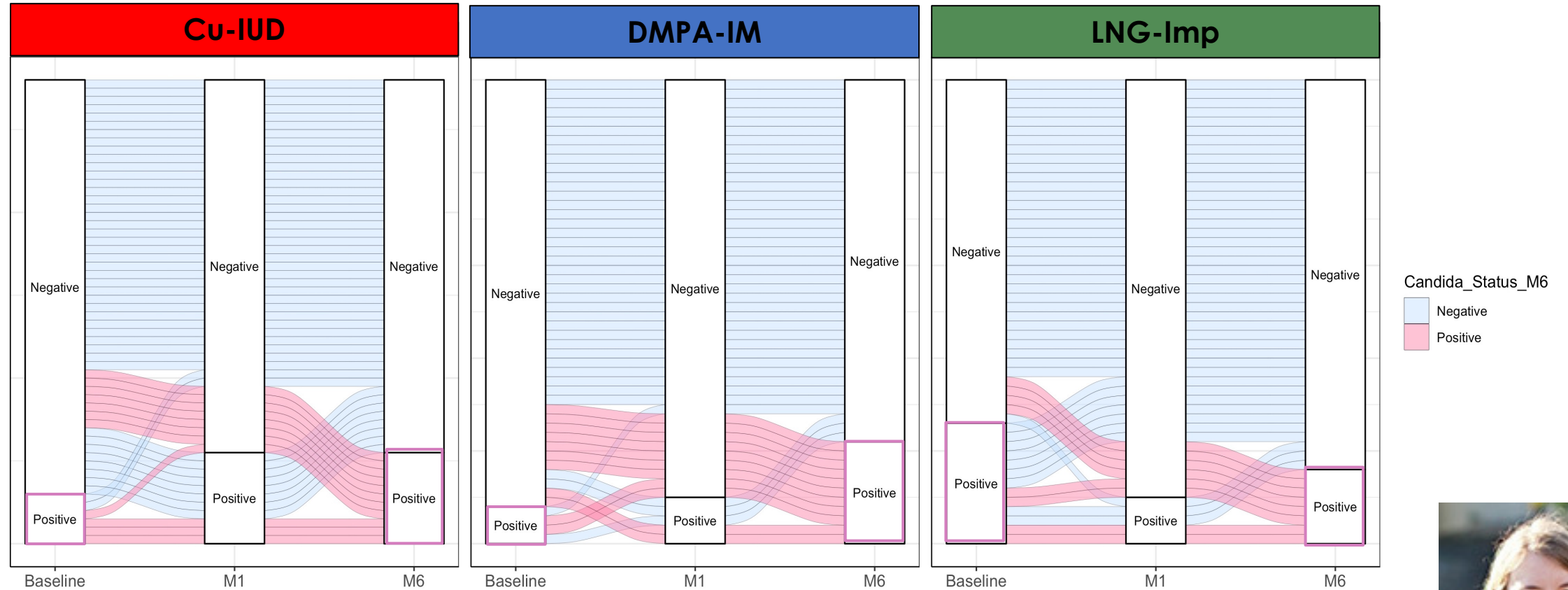
Th17 cell frequency strongly correlated with proteins involved in mucosal barrier integrity



No relationship between Th17 cell frequency and *Candida* detection

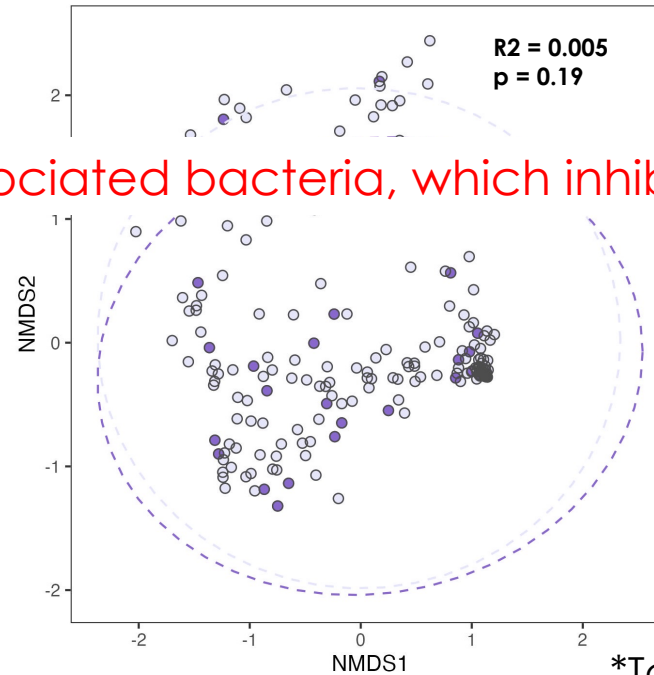
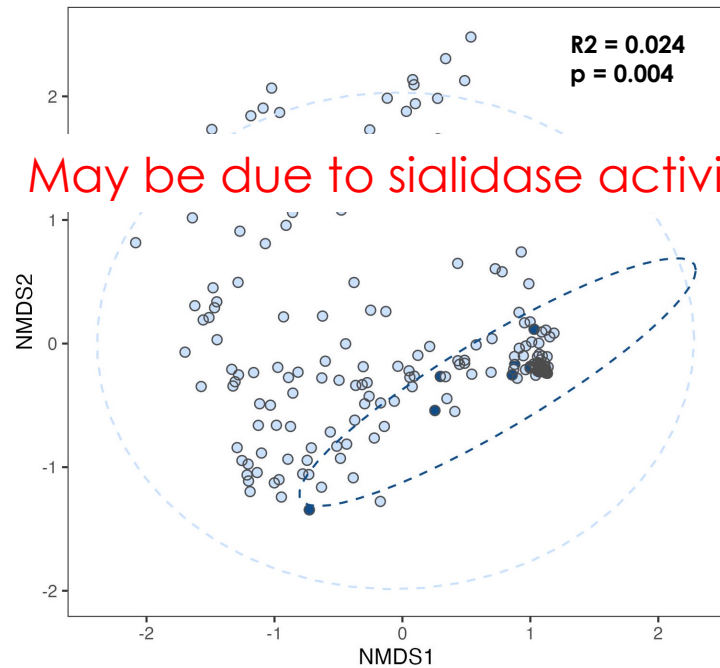
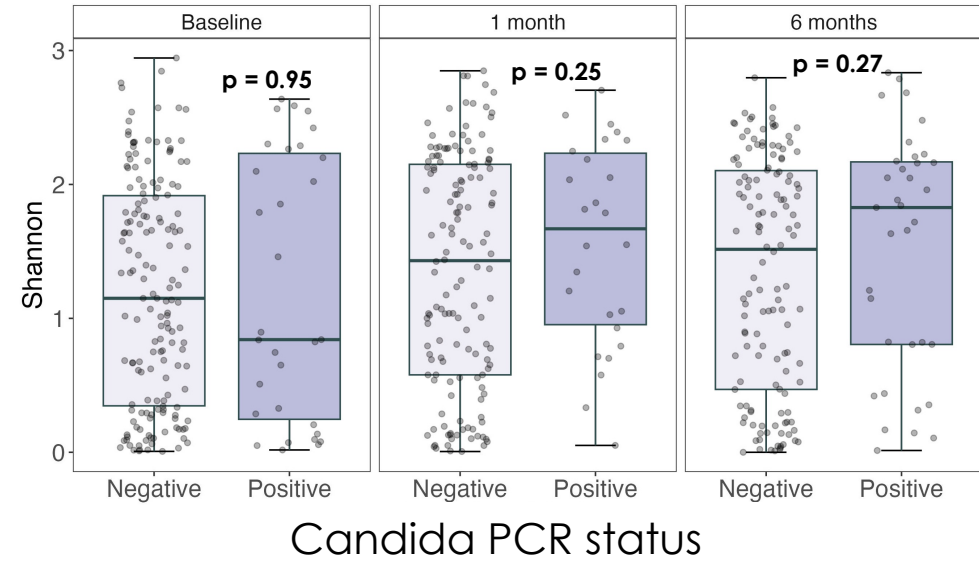
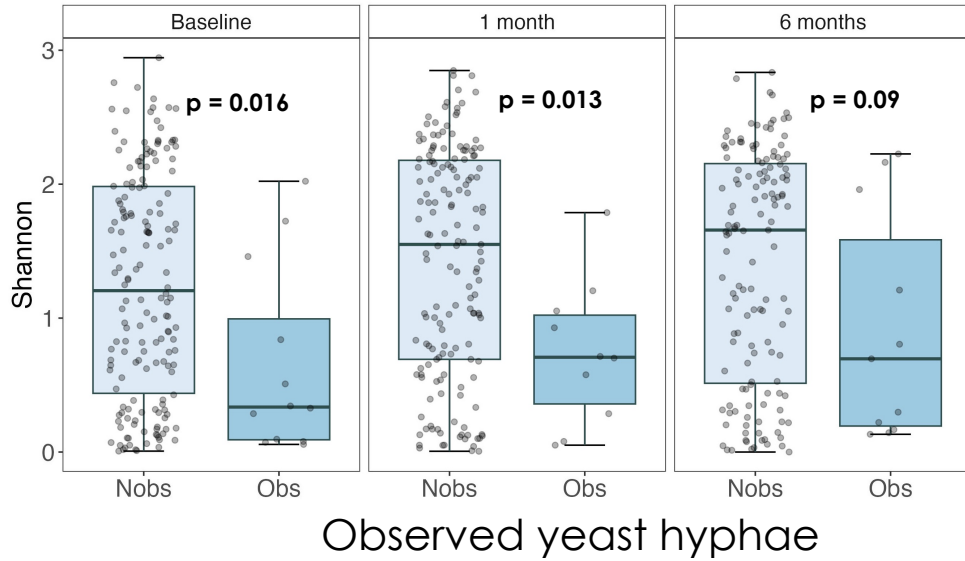


Contraceptive arm does not influence Candida detection by PCR



Mel Gasper, PhD

Candida hyphae presence related to bacterial diversity and composition

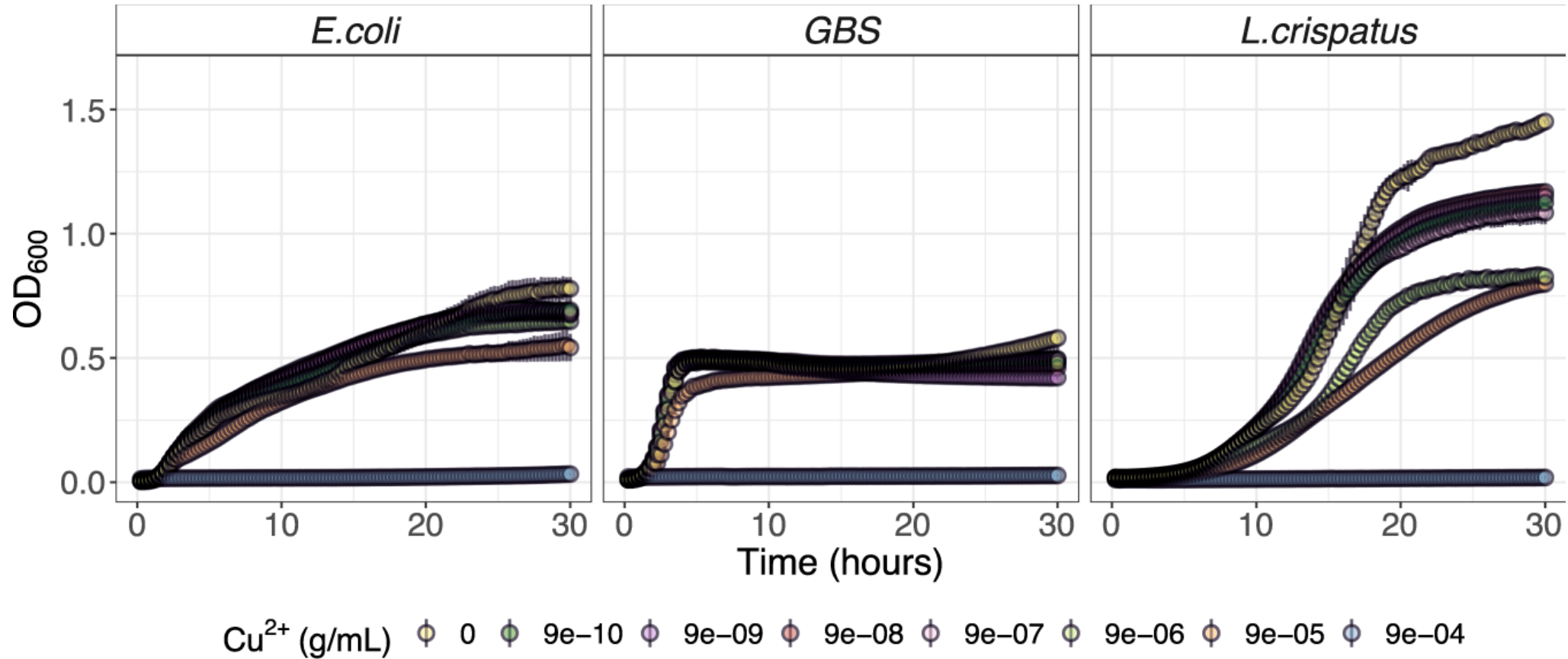


May be due to sialidase activity of BV-associated bacteria, which inhibits hyphae formation*

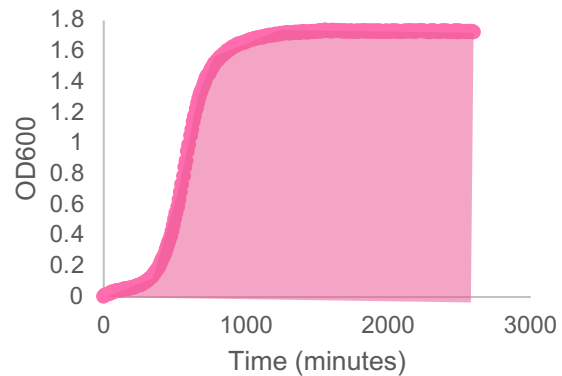
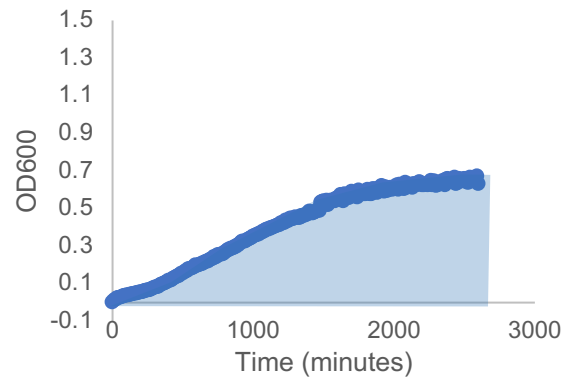
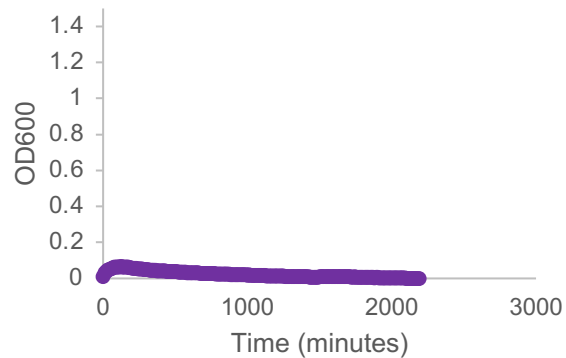
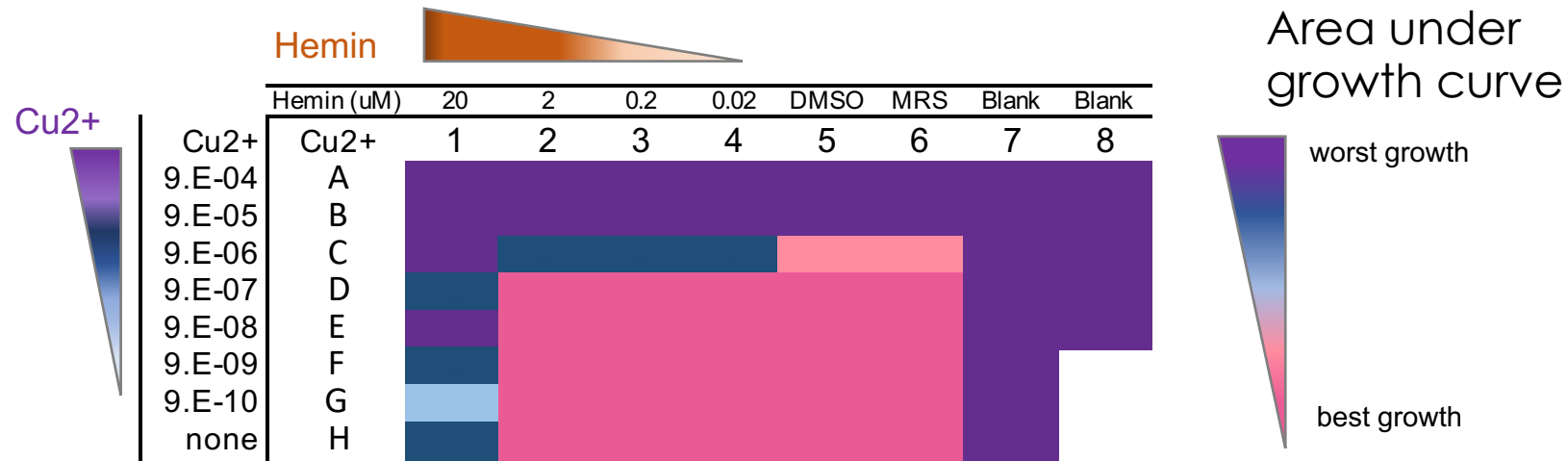
Mechanisms through which Copper-IUD could induce microbiota changes

- Copper could kill lactobacilli preferentially
 - Intracellular accumulation of cations
 - Induction of proviruses
- Copper-IUD induced bleeding could kill lactobacilli or encourage growth of BV-associated organisms
- Physical foreign body may itself alter microbiome
 - Test this in a future randomized trial with LNG-IUS

Copper sulphate inhibits *L. crispatus* growth in vitro in a dose-dependent manner



Copper is more toxic to lactobacilli than hemin (iron)

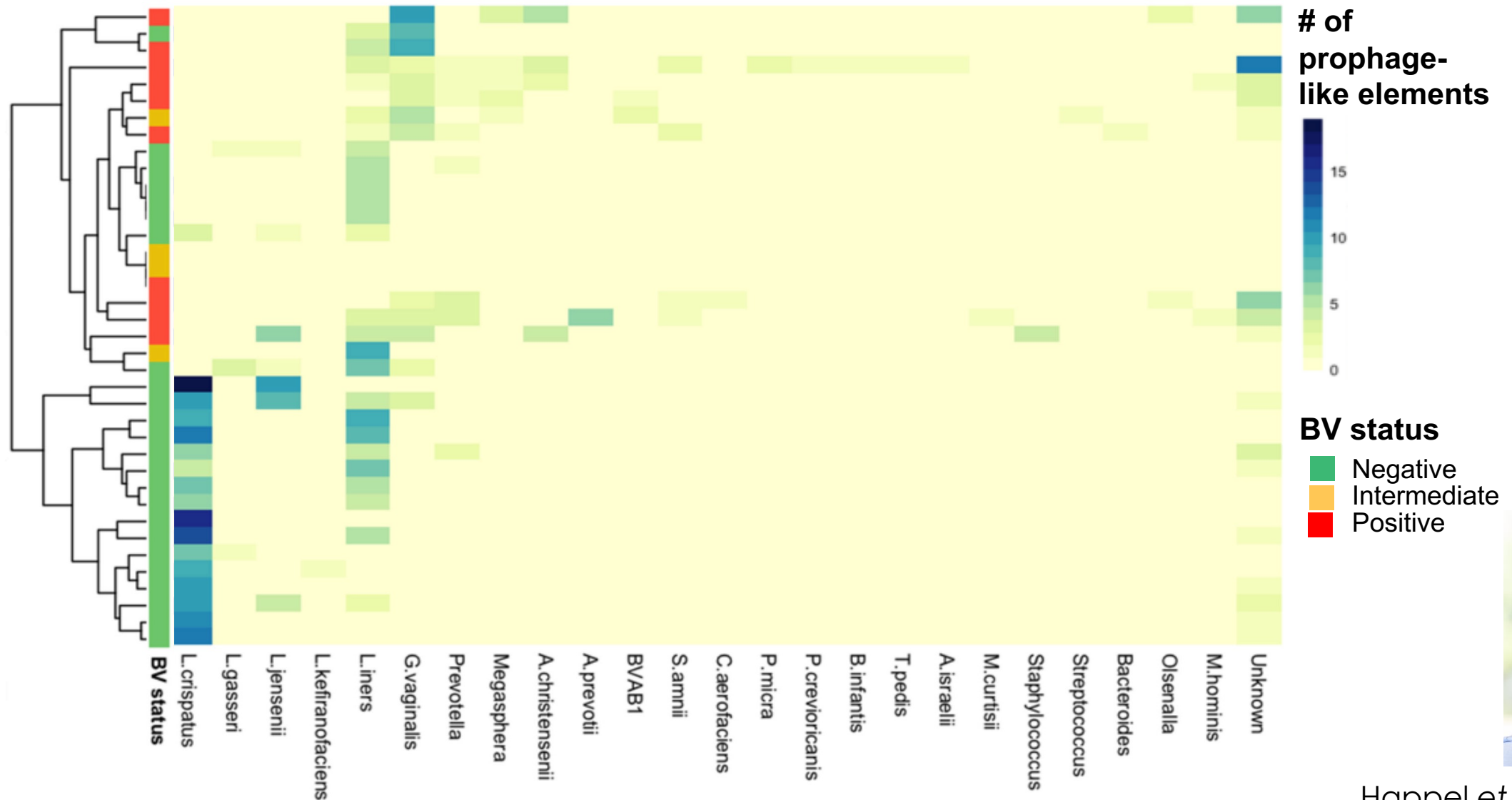


Sera Lee

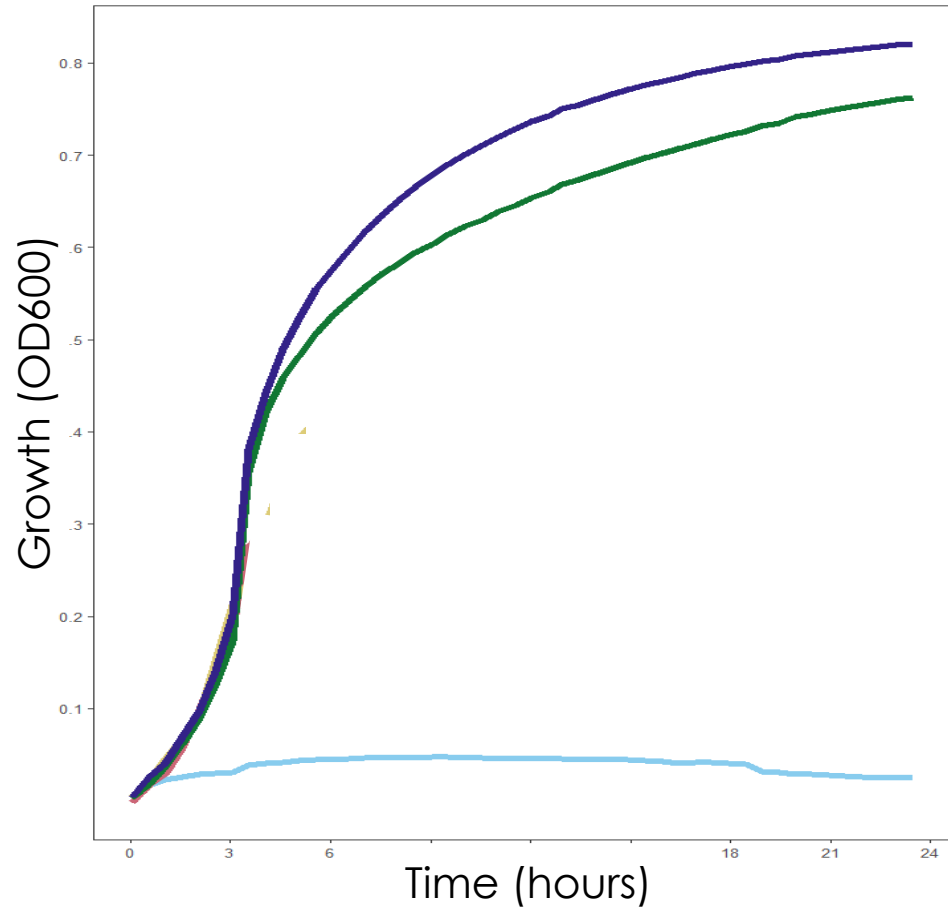


Mel Gasper, PhD

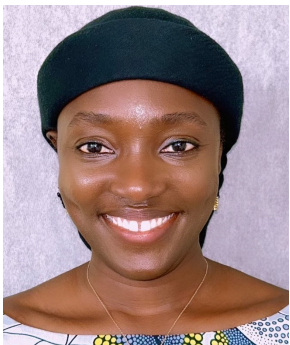
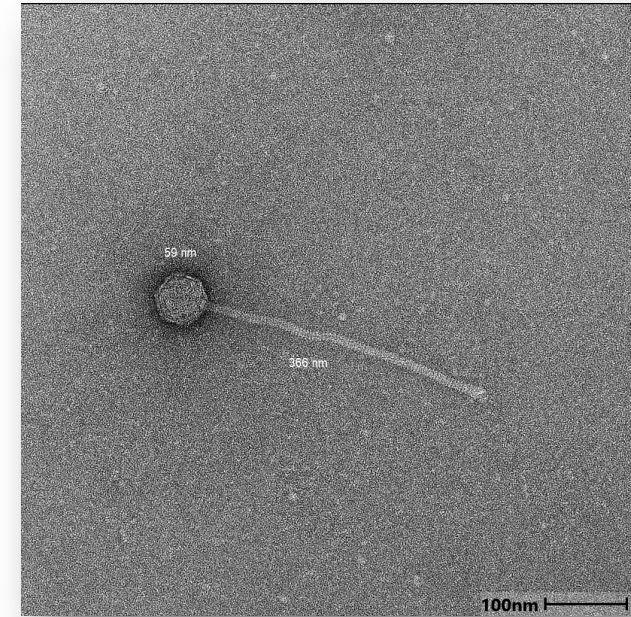
Lactobacilli in vaginal metagenomes have more prophage than other vaginal bacteria



Mitomycin induces phage from South African lactobacillus isolates



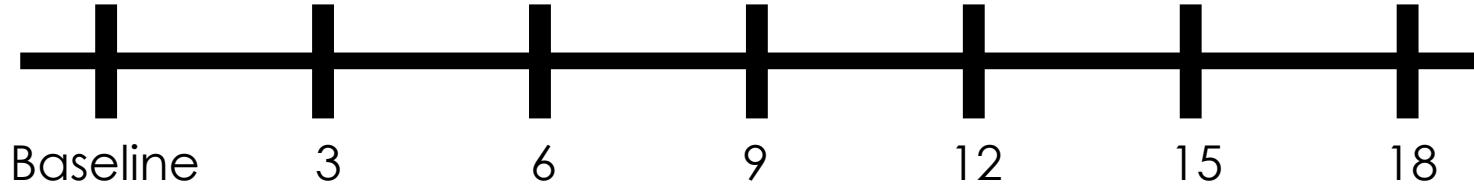
- Media alone
- L. crisp+Mitomycin
- L. crispatus



Adijat Jimoh

BMGF HPV Study

Time
[months]



Demographics
Sexual history
Reproductive history
Adherence

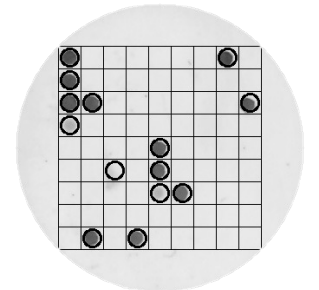


Sexual health

Bacterial vaginosis
Candidiasis
Pap smear



Measurement of vaginal
bacterial and fungal relative
abundances by 16s rRNA and
ITS gene sequencing



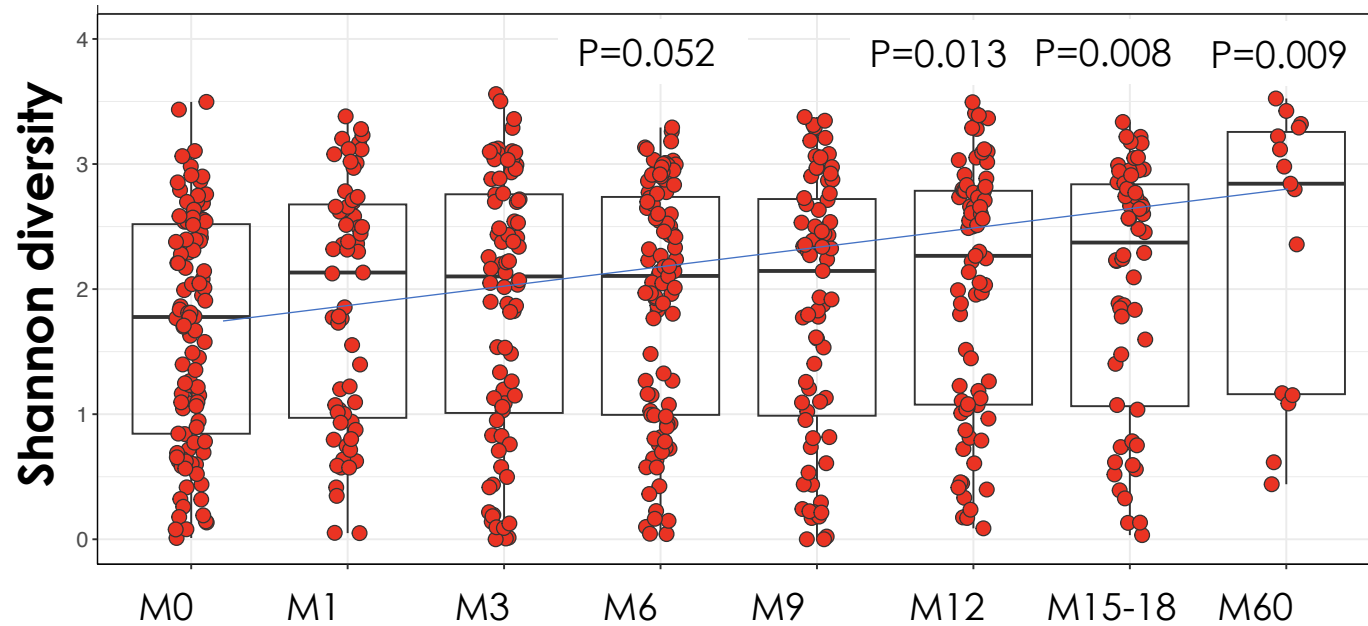
HPV Typing:
Direct Flow
CHIP Kit

- HPV is the most common STI globally and causes cervical cancer
- Observational data suggest relationship between HPV and vaginal microbiota

Baseline cohort characteristics by arm

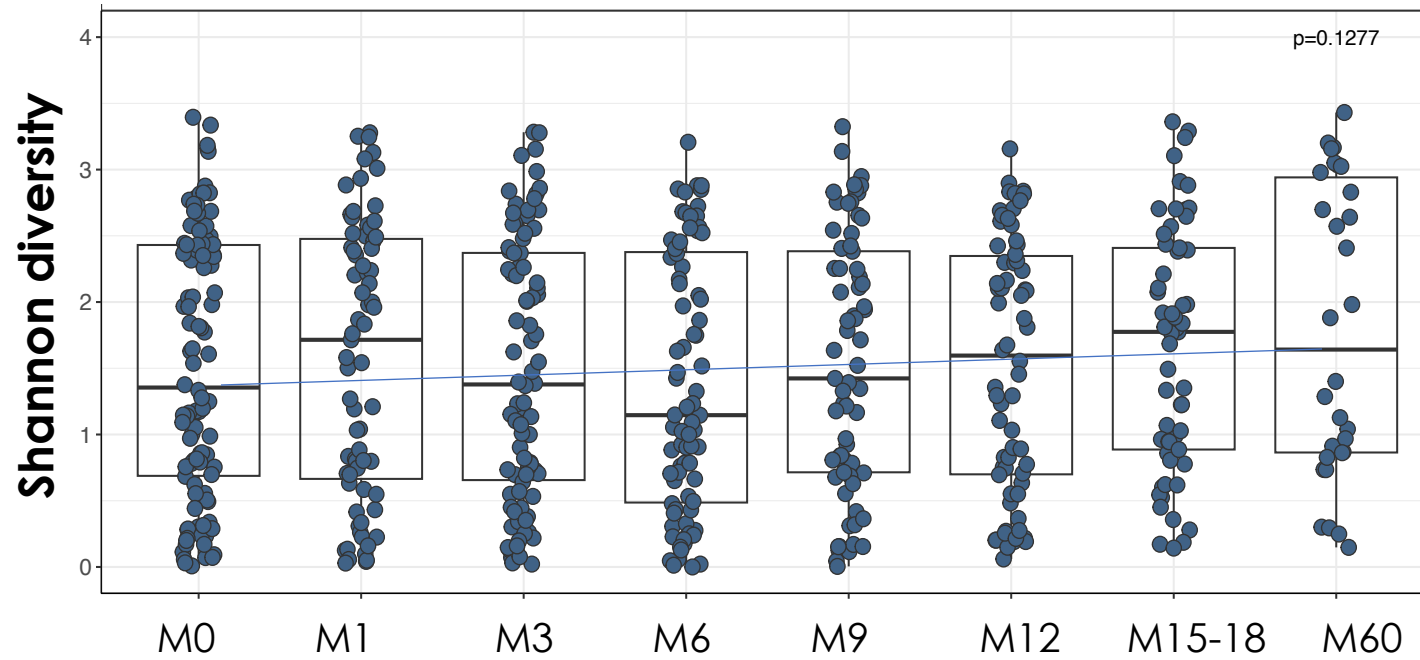
	DMPA (n=100)	IUD (n=107)	Implant (n=104)
Country = South Africa, n(%)	56 (56.0)	60 (56.1)	61 (58.7)
Median Age, years [IQR]	22 [21 - 27]	23 [20 - 28]	23 [20 - 26]
BMI > 30, n(%)	21 (21.2)	28 (26.2)	21 (20.4)
Primary education completed, n(%)	77 (77.0)	80 (74.8)	81 (77.9)
Married, n(%)	38 (38.0)	32 (29.9)	40 (38.5)
Any previous pregnancy, n(%)	89 (89.0)	92 (86.0)	88 (84.6)
Bacterial STIs, n(%)			
<i>N. gonorrhoea</i>	8 (8.0)	9 (8.4)	5 (4.9)
<i>C. trachomatis</i>	16 (16.0)	19 (17.8)	13 (12.7)
Any	21 (21.0)	24 (22.4)	16 (15.7)
Bacterial Vaginosis (Nugent \geq 7), n(%)	25 (26.9)	32 (30.8)	34 (36.2)
More than 1 sex partner, prior 3 months, n(%)	6 (6.0)	10 (9.3)	5 (4.8)
New sex partner, prior 3 months, n(%)	8 (8.0)	6 (5.6)	3 (2.9)
Any condomless sex, prior 3 months, n(%)	68 (68.0)	72 (67.3)	73 (70.2)

Effects of Cu-IUD on microbiota are sustained up to 5 years

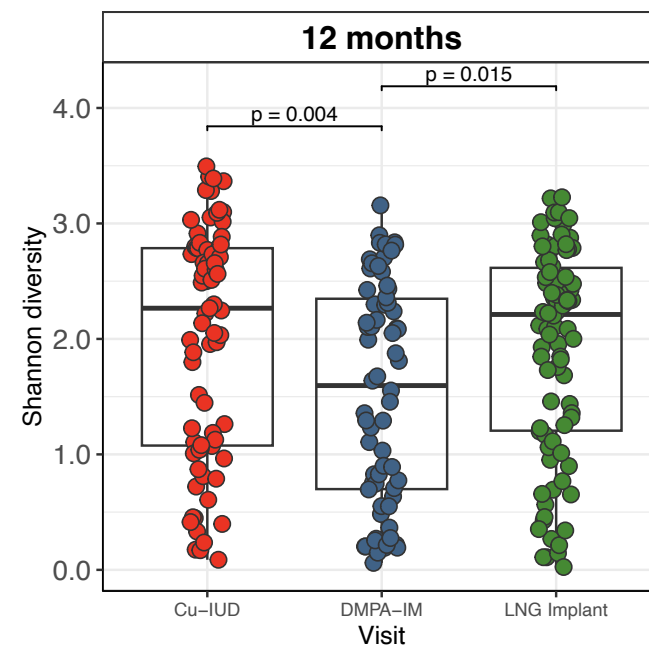
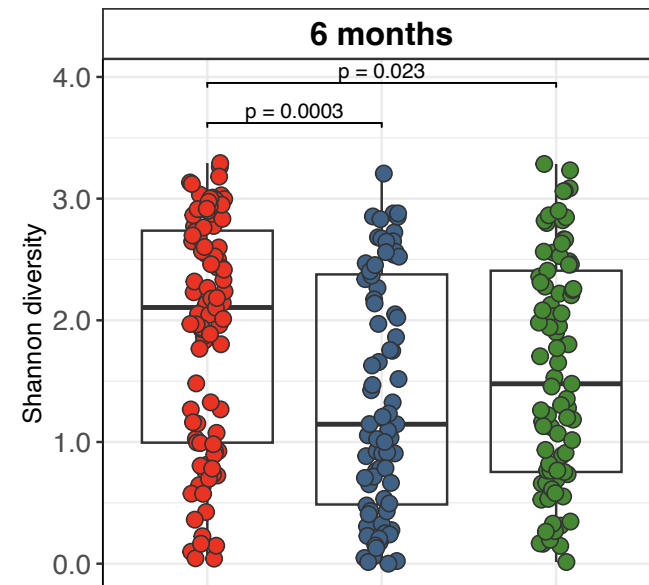
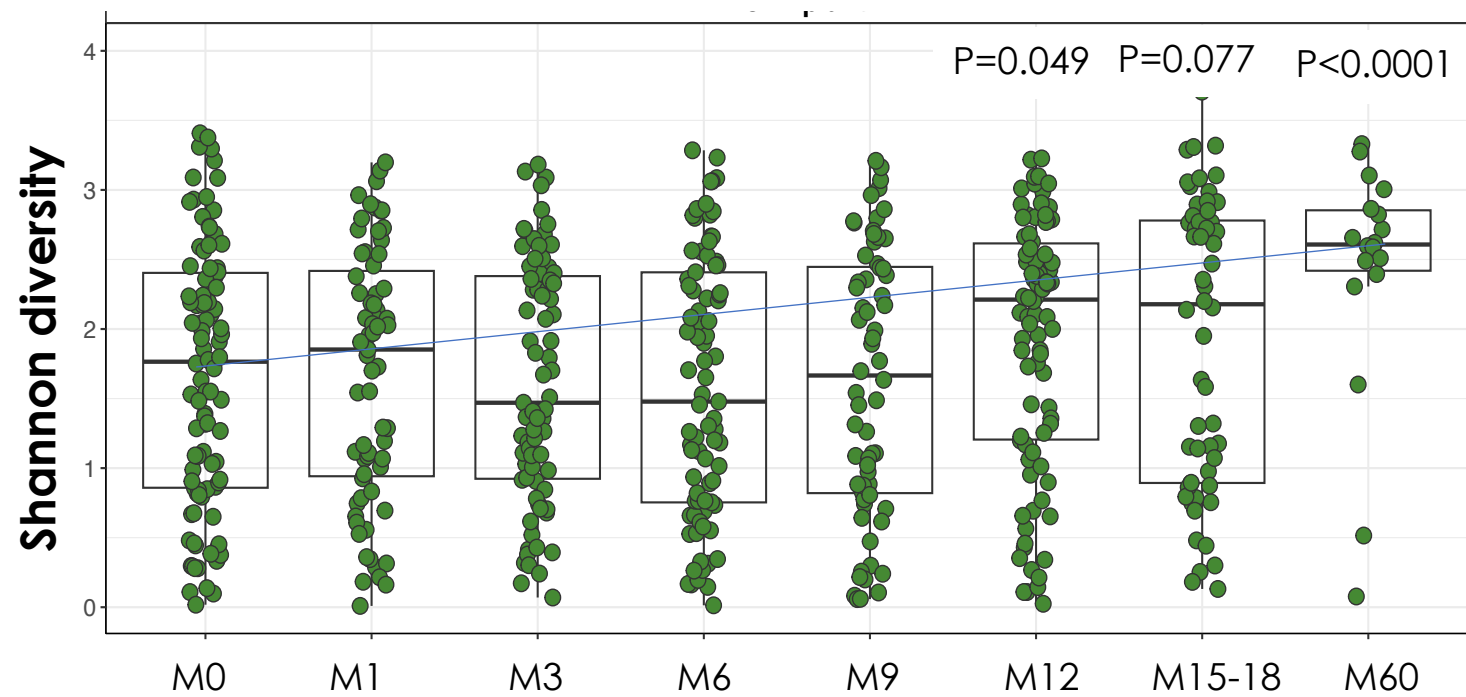


Smritee Dabee, PhD

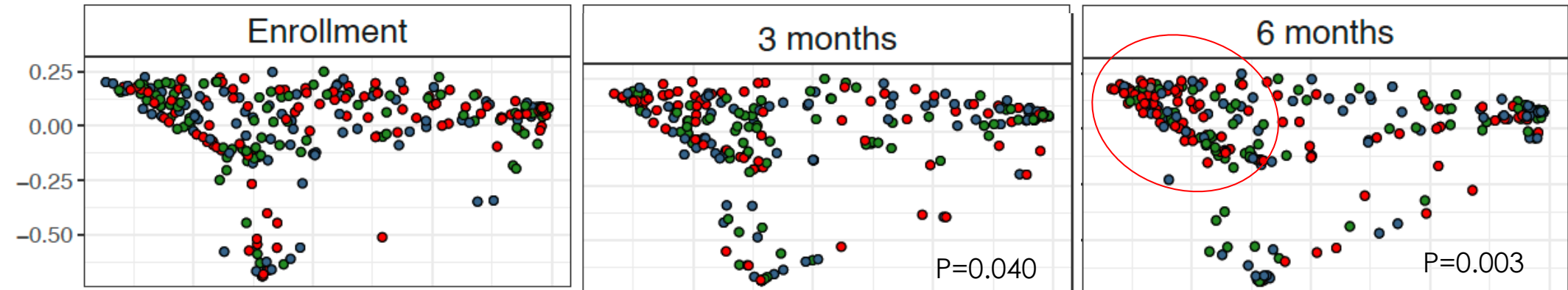
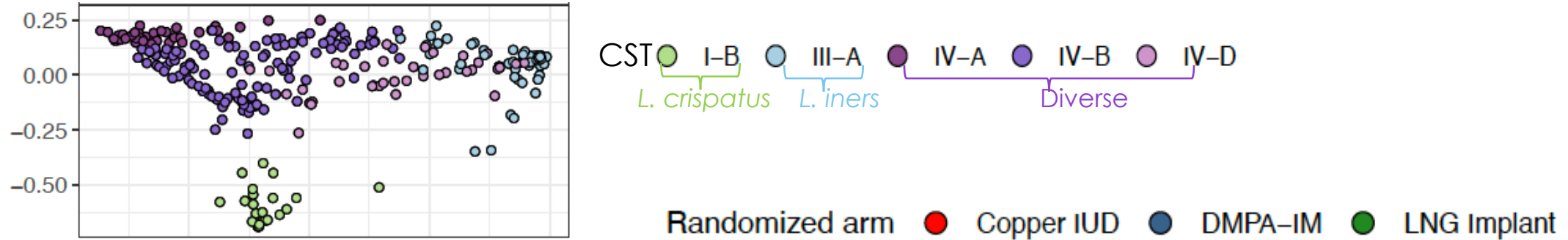
No effects of DMPA on microbiota up to 5 years



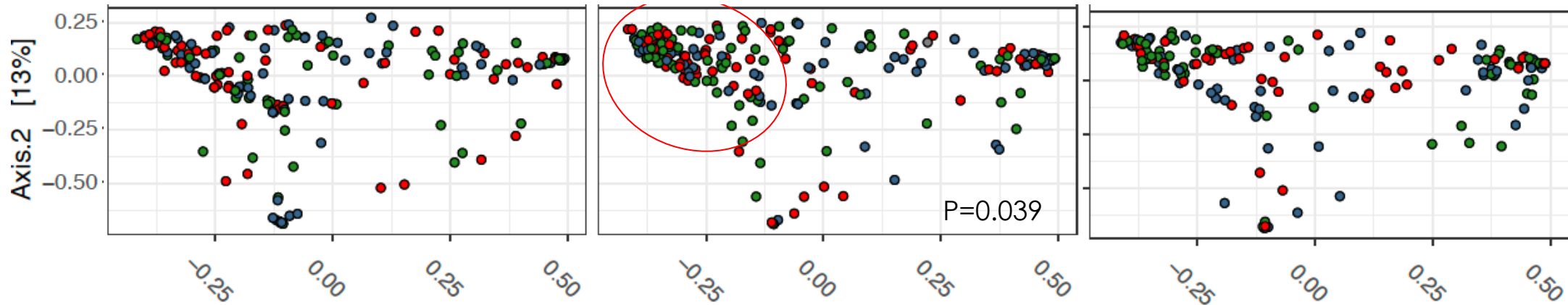
LNG implant increases vaginal microbial diversity with long term use



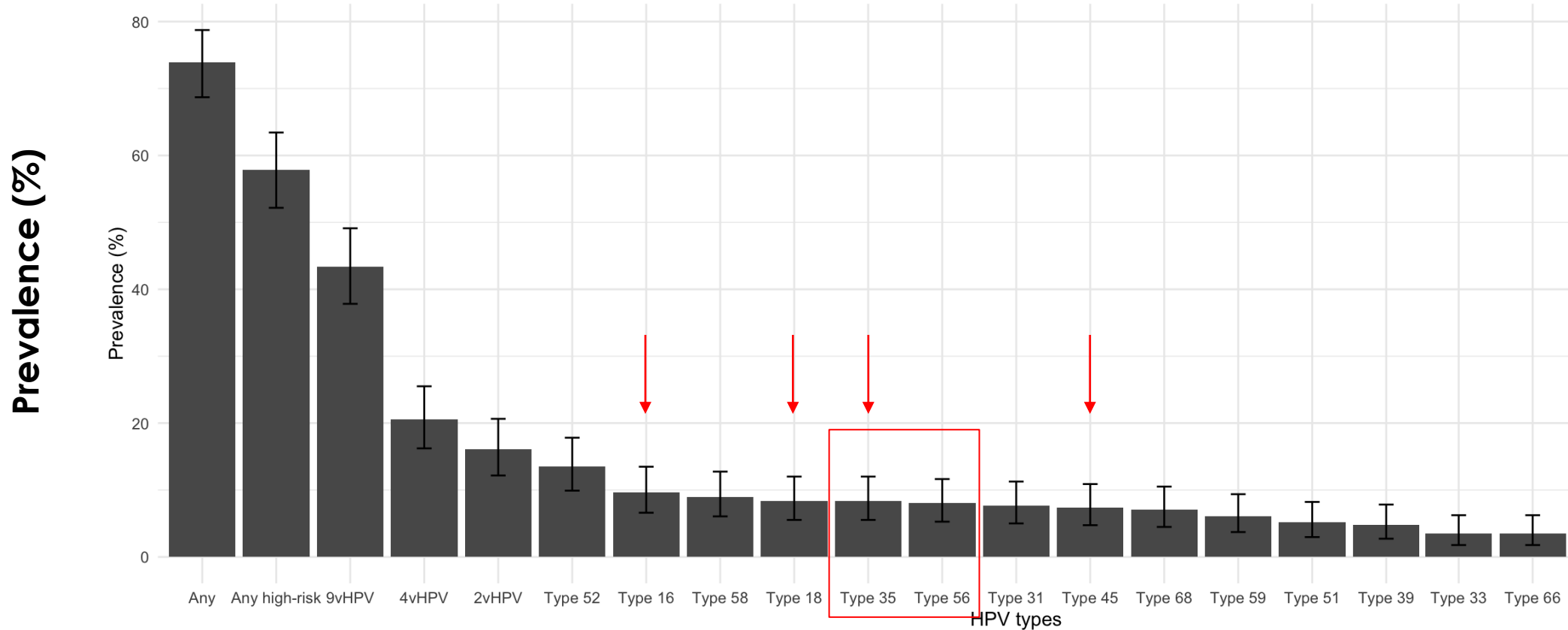
Arm explains a significant portion of community composition longitudinally



LNG-Implant **and** Cu-IUD induce a shift toward more diverse communities by 12 months of use

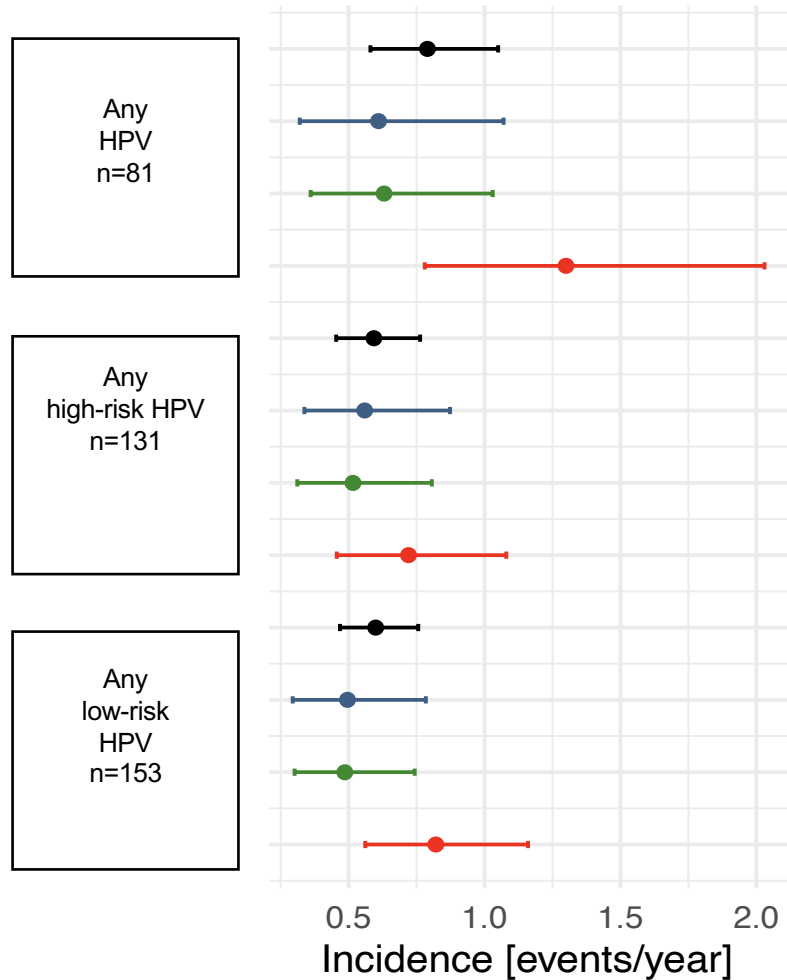


High prevalence of hr-HPV type

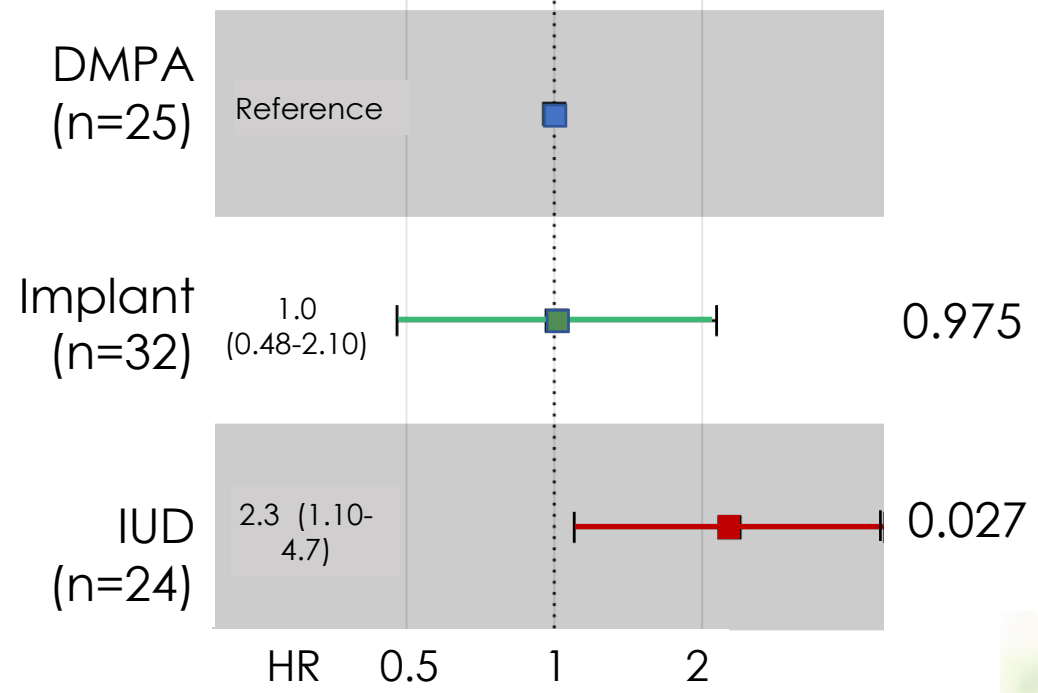


HPV-35 and -56 not targeted by any of the commercially available vaccines
HPV-16, -18, -35 and -45 are responsible for the majority of cancers worldwide

Cu-IUD users had higher incidence of any HPV incidence



Cox proportional Hazard ratio
Any HPV

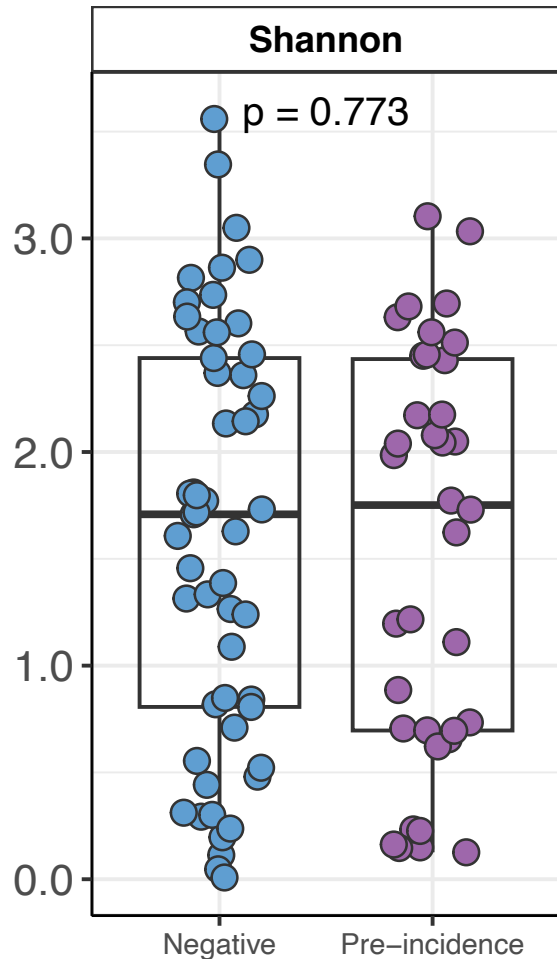


IUD vs LNG Implant HR 2.21 (1.13-4.44); $p = \mathbf{0.021}$

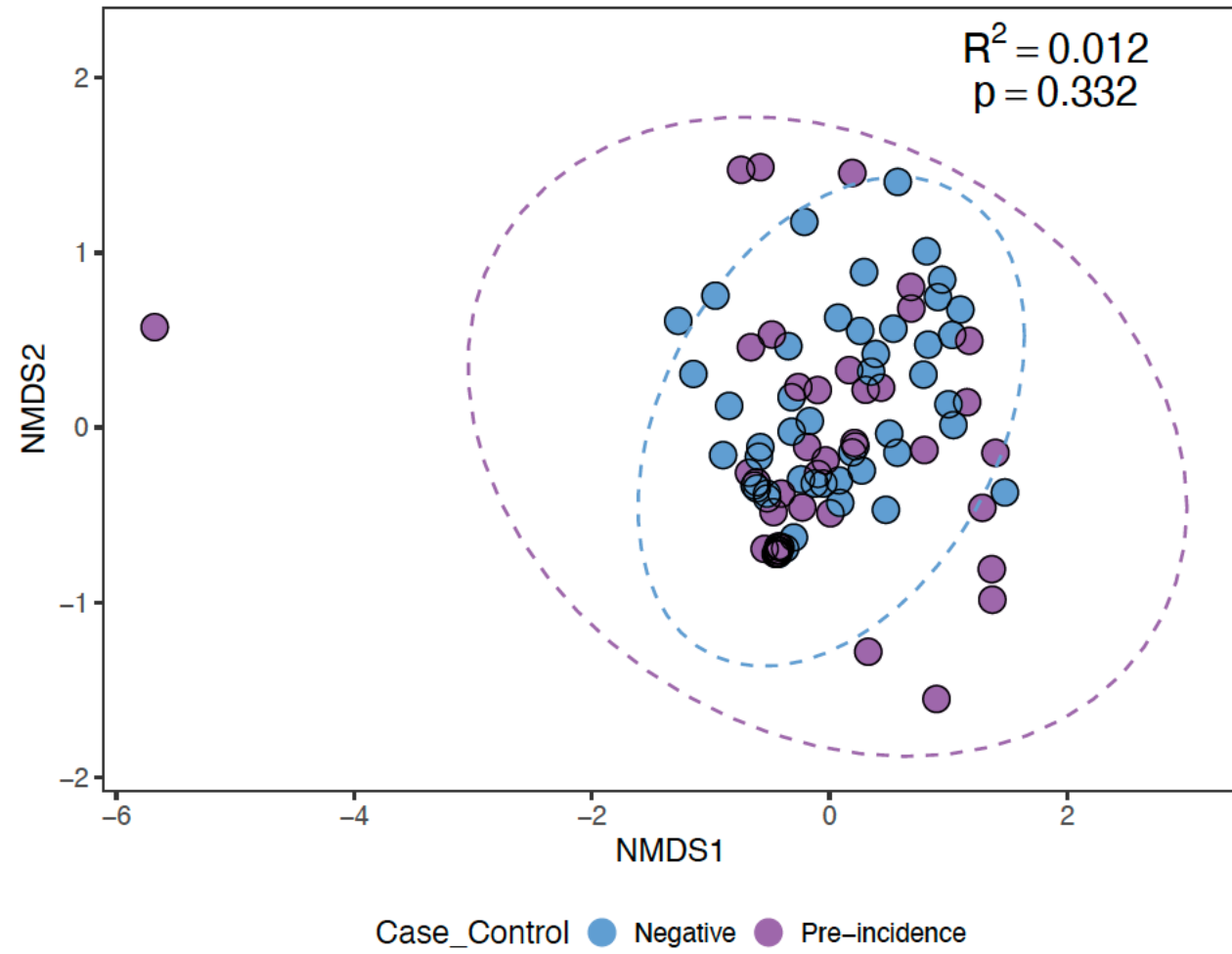


Anna Happel, PhD

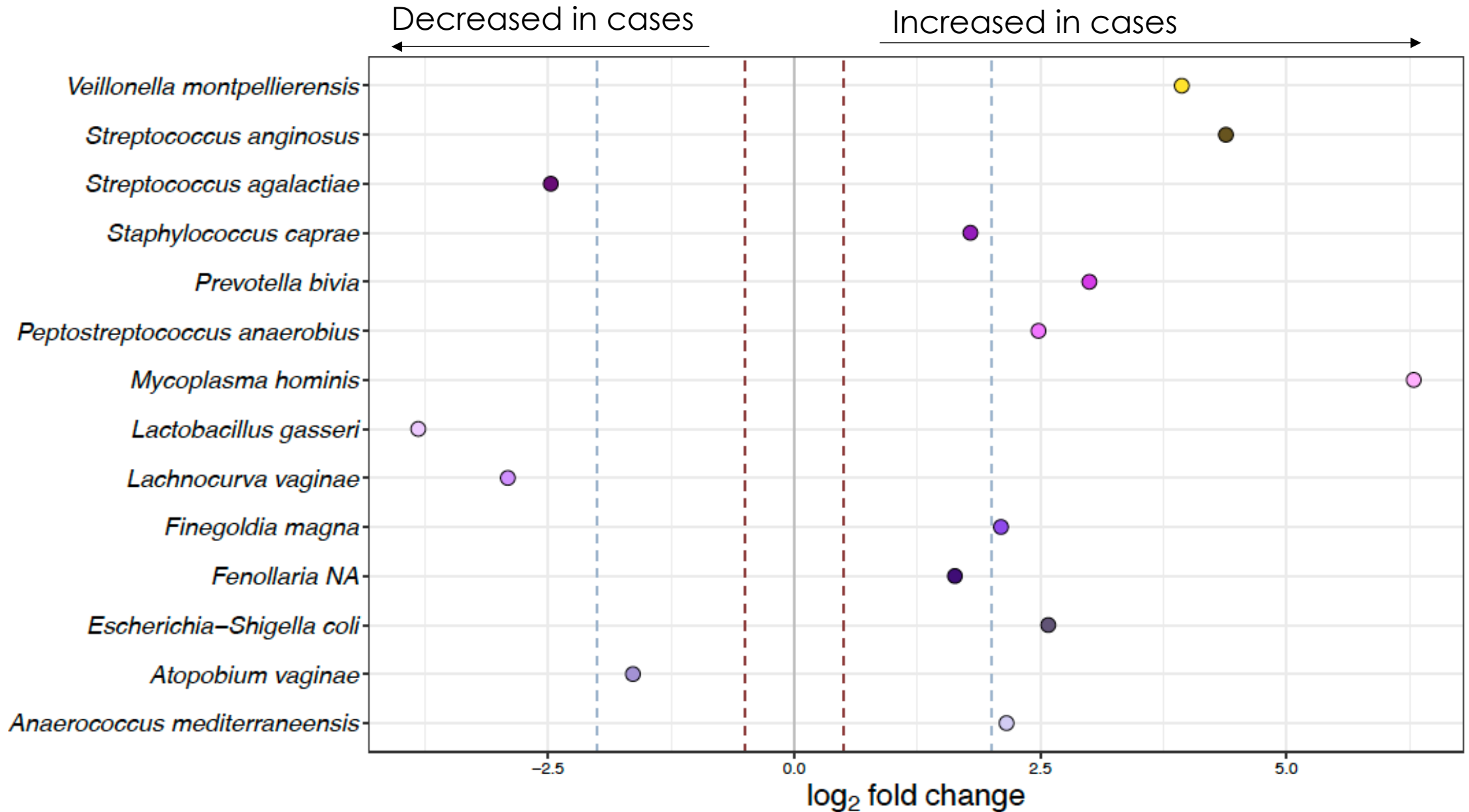
Vaginal microbial diversity does not play a role in HPV incidence



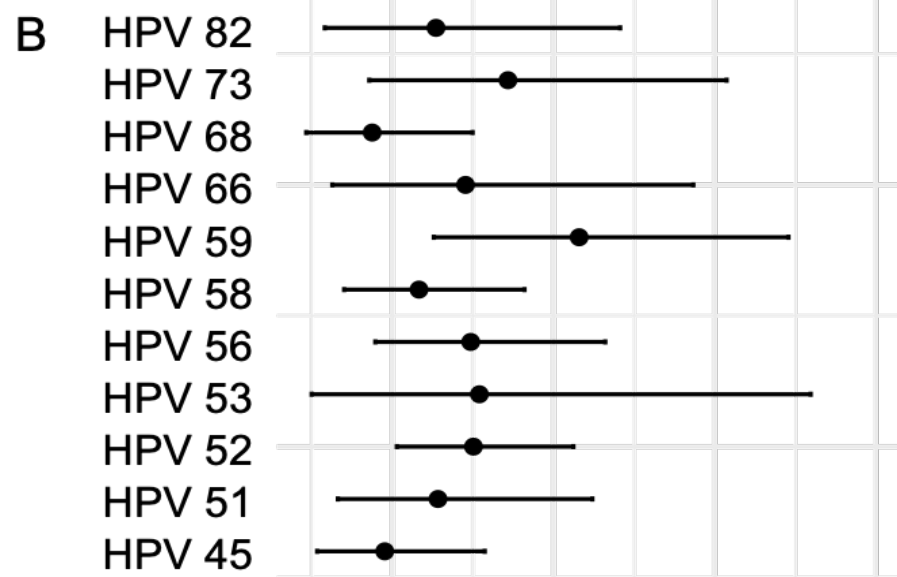
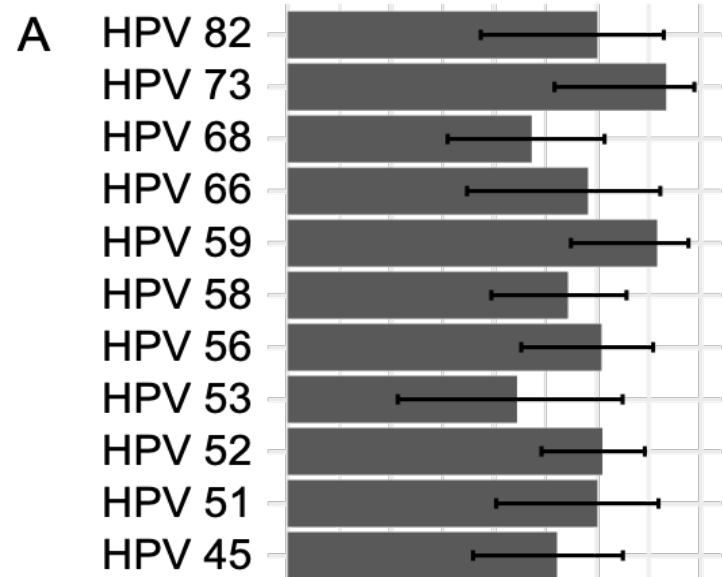
Any HPV incidence



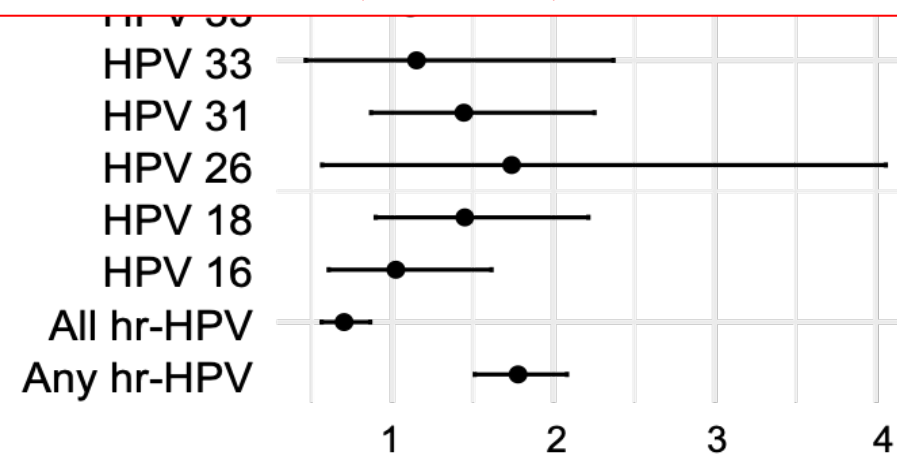
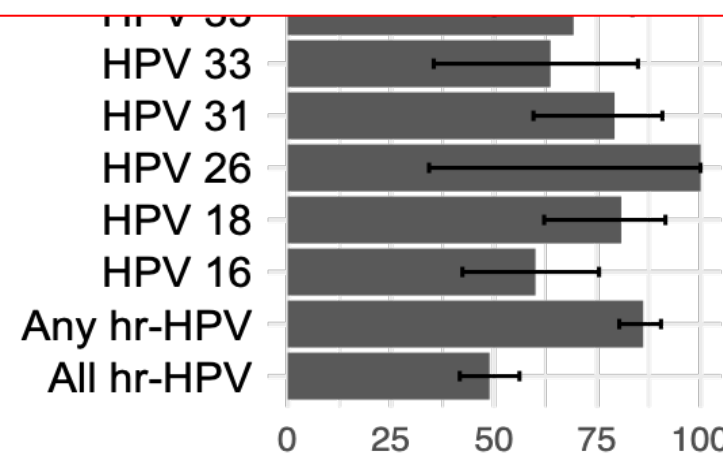
Specific microbes may play a role in HPV incidence



High proportion of women clearing hr-HPV infections 'naturally' over 18 months



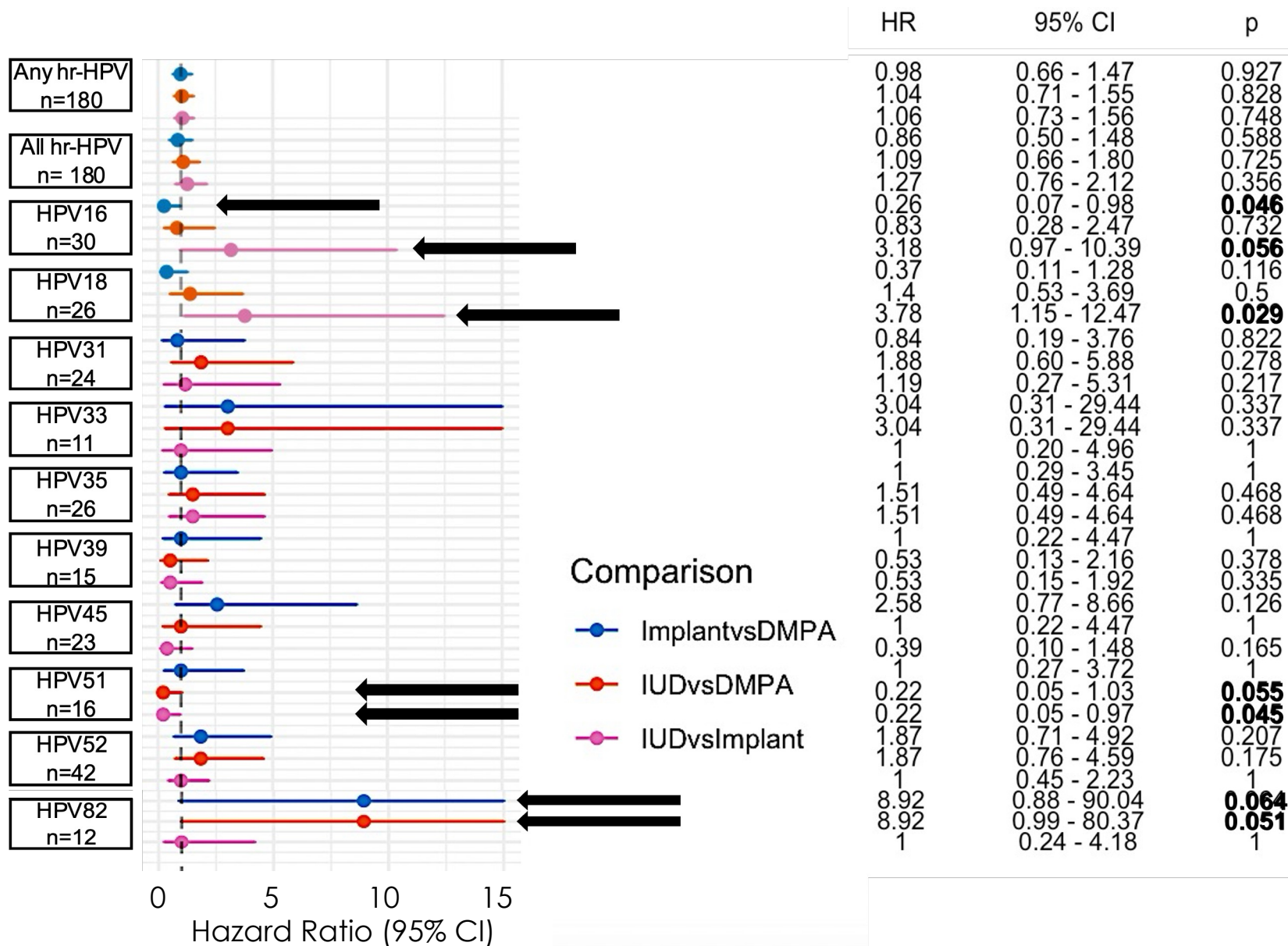
The lowest clearance rates were observed for HPV-35, HPV-16, HPV-45 and HPV-68



Proportion women who cleared (%)

Clearance rates (95% CI)

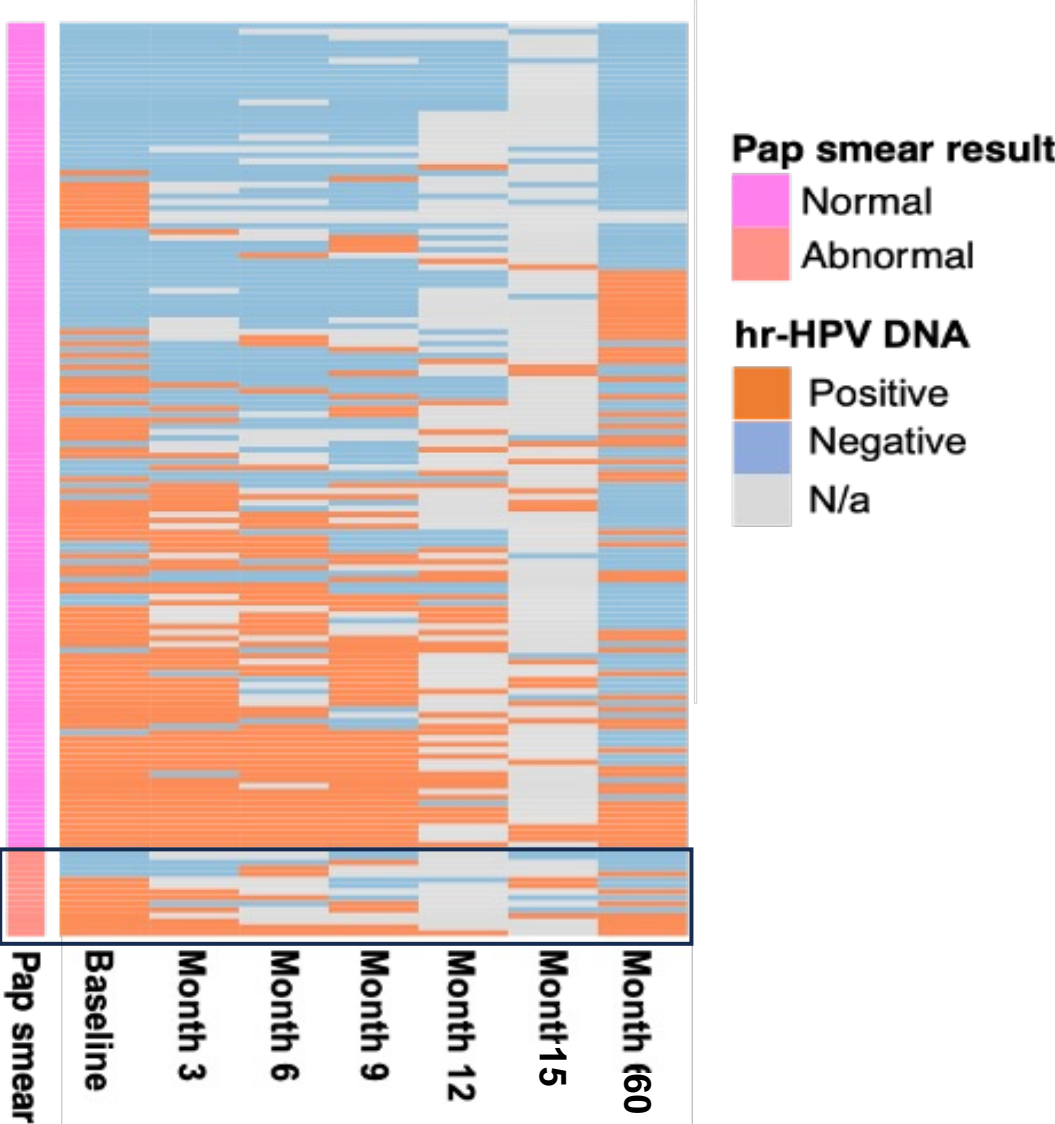
Clearance of hr-HPV between arms was type-specific



HPV-16:
Cu-IUD & DMPA
users more likely
to clear vs
Implant

HPV-18:
Cu-IUD users
more likely to
clear vs Implant

Few hr-HPV infections led to abnormal cytology at month 60



15 abnormal Pap smears
-Abnormal cytology in women never testing hr-HPV positive
-Normal cytology in women with persistent hr-HPV detection
-Possibly need African-specific HPV diagnostics

Summary

- The effects of Cu-IUD on vaginal microbiome persist up to 5 years
- LNG-Implant may also induce changes to vaginal microbiota after 1 year of use
- High-risk HPV genotypes are common in east and southern Africa. African-specific HPV vaccines are vital to reduce cervical cancer.
- HPV incidence was higher in women assigned to Cu-IUD compared to those assigned to DMPA and LNG-implant.
- This increased incidence does not seem to be related to Cu-IUD effects on the microbiota
 - Role of specific microbes could be explored
- Since Cu-IUD is the only non-hormonal, reversible, long term contraceptive option for many women, requires further inquiry.

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