Pooling for SARS-CoV-2 testing: the Ghana Experience

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Ghana's experience with 'pooling'

Why we decided to 'pool'

- 1. We were going to be receiving in excess of 3,000 samples/day
- 2. Save reagents and be able to have results in realistic time
- 3. Our prevalence was low (<2%), this means that after testing 100 samples, we will get about 1 being positive

'Pooling' is not a new method

- ❖ Troop education and avian influenza surveillance in military barracks in Ghana, 2011 (Odoom et al., BMC Public Health, 2012)
- Authors sampled a total of 680 birds from 102 households.

A total of 824 samples (tracheal and cloacal swabs) were pooled into 94 pools and tested for the presence of influenza virus. All pools were negative for influenza virus.

Validation of 'pooling' at NMIMR

Pool 1			Pool 2		
Sample ID	Previous results	Results after	Sample ID	Previous results	Results after
		unpooling			unpooling
nCoV-Ct-153	Negative	Negative	nCoV-Ct-16	Negative	Negative
nCoV-Ct-154	Negative	Negative	nCoV-Ct-17	Negative	Negative
nCoV-Ct-155	Negative	Negative	nCoV-Ct-18	Negative	Negative
nCoV-Ct-156	Negative	Negative	nCoV-Ct-19	Negative	Negative
nCoV-Ct-157	Negative	Negative	nCoV-Ct-20	Negative	Negative
nCoV-Ct-158	Negative	Negative	nCoV-Ct-21	Negative	Negative
nCoV-Ct-159	Negative	Negative	nCoV-Ct-22	Negative	Negative
nCoV-Ct-160 (pos)	*29.28; **31.09	*32.06; **36.13	nCoV-Ct-23	Negative	Negative
nCoV-Ct-161	Negative	Negative	nCoV-Ct-24 (pos)	*24.21; **27.56	*26.83; **31.85
nCoV-Ct-162	Negative	Negative	nCoV-Ct-25	Negative	Negative

Key: * Cycle threshold values for Open Reading Frame (ORF) 1ab; ** Cycle threshold values for Nucleocapsid (N) gene



Volumes of samples 'pooled'

RNA Extraction Kit	Volume of sample in a pool (μL)	Total volume of sample/pool (μL)
QIAamp Viral RNA Minikit	14	140
Beaver Nucleic Acid Extraction kit	20	200
Zymo Quick-RNA Miniprep kit	25	250
RNeasy kit	14	140
DAAN Gene RNA Extraction kit	20	200
Viral Nucleic Acid Extraction kit II (Geneaid)	20	200
NX-48S Viral RNA Extraction kit (Genolution)	20	200

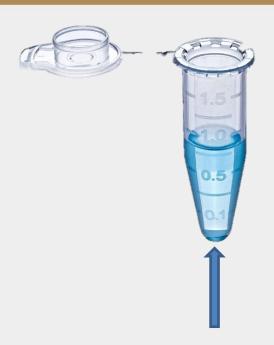


What was done in Ghana?

We 'pooled' 5/10 samples together to make one sample



What was done in Ghana



5/10 samples combined as one sample, mix very well (vortex)



RNA extraction



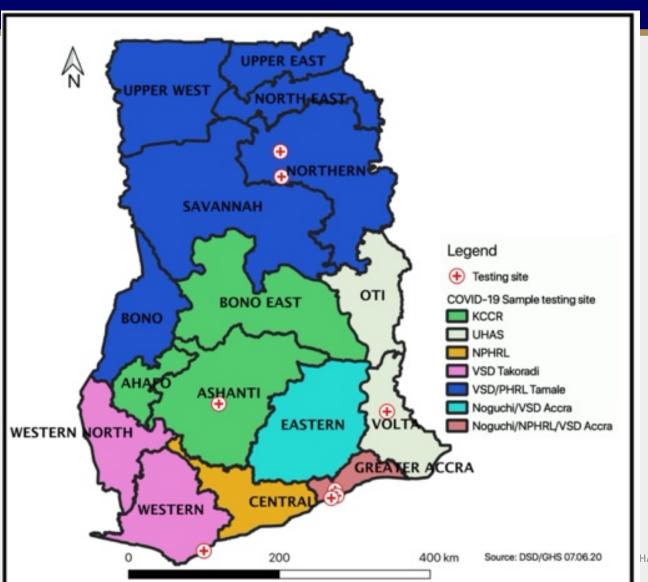


Real time PCR

Positive pools are 'unpooled' (samples are tested individually)



Which labs were pooling....



All labs doing COVID-19 testing in Ghana; including peripheral labs

NMIMR (main lab that started pooling)
KCCR (main lab)
UHAS (peripeheral lab)
NPHRL (peripeheral lab)
VSD/PHRL Tamale (peripeheral lab)
VSD Accra (peripeheral lab)



Why we stopped 'pooling'

❖Our prevalence increased (from <2% to approximately 10%), number of positive samples per number of tests started increasing

- ❖ Number of positive pools increased and we were dissolving a lot of pools (80 out of 92 pools tested; 87% of pools)
- When 50% of pools are positive; consider not pooling
- Results were delaying because we had to dissolve a large number of pools

What we achieved...

- From April to June, we were able to test 105, 464
- ❖ Pooling increased our testing capacity from 1,000 samples/day to about 10,000 samples/day (saving approximately 250,000 USD)

'Pooling' does not affect sensitivity of rRT-PCRs adversely

'Pooling' saves resources (especially for resource limited areas) and time

