Twitter Thread by Francisco de Asis



Francisco de Asis
@franciscodeasis

[Thread] Pangolin CoV... or Bat CoV in pangolins samples?
TLDR: A researcher of the team that sequenced the pangolins samples had taken samples in the mine of RaTG13 and in the place where RmYN02 was collected, also catched bats in Yunnan. Probably contaminated pangolin samples

@Daoyu15 was the first to note that the raw sequence reads of the pangolins “contained unexpected reads and was in serious risk of contamination” in early June 2020. But now we can explain how it was possible to happen this in Guangdong https://t.co/SbBi7JXX5A

Jin-Ping Chen, the corresponding author of the first paper of the pangolins, and LiBiao Zhang, who has been sampling the mine of RaTG13 and the place in Mengla where RmYN02 was collected, are close colleagues in GIABR & GIZ
https://t.co/Mf0AeRatge
https://t.co/C444TLT6JO



LiBiao Zhang was probably in the mine on 27-Jul-12 with HKU team (he is co-author in Lau et al. (2017)) and a week later on 06-Aug-12 with WIV in their 1st visit. Surprisingly, he is co-author in the AdV isolates paper, but not credited in Ge et al. (2016)

https://t.co/ZNW6sd976T

Viruses	Accessior	Genera	Spe	Cite	Group	sa	custon	ci	ci	ca	cave nam	cave prov	cave region	collecti	collec
Bat coronavirus MLHJC4	KU182954	Alphacoronavirus		Xu et al. (2016)	PLA		MLHJC4	CN	YN	MLHJ	Menglian	Yunnan	Southern		Jul-12
Bat coronavirus MLHJC6	KU182968	Alphacoronavirus		Xu et al. (2016)	PLA		MLHJC6	CN	YN	MLHJ	Menglian	Yunnan	Southern		Jul-12
Bat coronavirus MLHJC8	KU182969	Alphacoronavirus		Xu et al. (2016)	PLA		MLHJC8	CN	YN	MLHJ	Menglian	Yunnan	Southern		Jul-12
Bat coronavirus MLHJC22	KU182970	Alphacoronavirus		Xu et al. (2016)	PLA		MLHJC22	CN	YN	MLHJ	Menglian	Yunnan	Southern		Jul-12
Bat coronavirus MLHJC34	KU182971	Alphacoronavirus		Xu et al. (2016)	PLA		MLHJC34	CN	YN	MLHJ	Menglian	Yunnan	Southern		Jul-12
Bat coronavirus MLHJC35	KU182963	Betacoronavirus	SARSr	Xu et al. (2016)	PLA		MLHJC35	CN	YN	MLHJ	Menglian	Yunnan	Southern		Jul-12
Mammalian orthoreovirus MRV-XN3662	KT444451	Orthoreovirus		Yang et al. (2015)	WIV	3662		CN	HB	XN	Xianning	Hubei	Far from Yunnan	19-Jul-12	Jul-12
Rousettus leschenaultii bocaparvovirus 1 RoI-BtBoV1_56C_ML_Y	MF682925	Dependoparvovirus		Lau et al. (2017)	HKU+GIABR+CDC		56C	CN	YN	ML	Mengla	Yunnan	Southern	24-Jul-12	Jul-12
Rhinolophus pusillus adeno-associated virus 1 Rp-BtAAV1_34C_	MF682926	Dependoparvovirus		Lau et al. (2017)	HKU+GIABR+CDC		34C	CN	YN	MJ	Mojiang	Yunnan	Southern	27-Jul-12	Jul-12
Rhinolophus pusillus bocaparvovirus 1 Rp-BtBoV1_48C_MJ_YN_2	MF682922	Dependoparvovirus		Lau et al. (2017)	HKU+GIABR+CDC		48C	CN	YN	MJ	Mojiang	Yunnan	Southern	27-Jul-12	Jul-12
Rhinolophus pusillus bocaparvovirus 2 Rp-BtBoV2_83C_MJ_YN_2	MF682923	Dependoparvovirus		Lau et al. (2017)	HKU+GIABR+CDC		83C	CN	YN	MJ	Mojiang	Yunnan	Southern	27-Jul-12	Jul-12
Mammalian orthoreovirus RpMRV-YN2012	KM087105, KM(	Orthoreovirus		Wang et al. (2015b)	CDC+Oth			CN	YN	?	YN Unknown	Yunnan	YN Unknown	01-Aug-12	Aug-12
Rat bocavirus MKRN/010812/1-RV	KX901832	Bocaparvovirus						CN	HK			Hong Kong	Far from Yunnan	01-Aug-12	Aug-12
Bat mastadenovirus WIV12	KT698856	Mastadenovirus		Tan et al. (2017)	WIV+GIABR+YIEDC		WIV12	CN	YN	MJ	Mojiang	Yunnan	Southern	06-Aug-12	Aug-12
Bat mastadenovirus WIV13	KT698852	Mastadenovirus		Tan et al. (2017)	WIV+GIABR+YIEDC		WIV13	CN	YN	MJ	Mojiang	Yunnan	Southern	06-Aug-12	Aug-12
WIV15	Not found	Unknown			WIV		WIV15	CN	?	?	Unknown	Unknown	Unknown		
Miniopterus bat coronavirus BtCoV/3709	KP876516, KU5	Alphacoronavirus	1	Ge et al. (2016)	WIV	3709		CN	YN	TG	Tongguan	Yunnan	Southern	06-Aug-12	Aug-12
Miniopterus bat coronavirus BtCoV/3710	KP876518, KU5	Alphacoronavirus	1	Ge et al. (2016)	WIV	3710		CN	YN	TG	Tongguan	Yunnan	Southern	06-Aug-12	Aug-12
Rhinolophus bat coronavirus BtCoV/3716	KP876509, KU5	Alphacoronavirus	1	Ge et al. (2016)	WIV	3716		CN	YN	TG	Tongguan	Yunnan	Southern	06-Aug-12	Aug-12
Hipposideros bat coronavirus BtCoV/3723 HBtCoV/3723	KP876529, KU5	Alphacoronavirus	HKU10	Ge et al. (2016)	WIV	3723		CN	YN	TG	Tongguan	Yunnan	Southern	06-Aug-12	Aug-12
Miniopterus bat coronavirus BtCoV/3728-1	KP876521	Alphacoronavirus	1	Ge et al. (2016)	WIV	3728	1	CN	YN	TG	Tongguan	Yunnan	Southern	06-Aug-12	Aug-12
Miniopterus bat coronavirus BtCoV/3728-2	KP876522	Alphacoronavirus	HKU8	Ge et al. (2016)	WIV	3728	2	CN	YN	TG	Tongguan	Yunnan	Southern	06-Aug-12	Aug-12
Miniopterus bat coronavirus BtCoV/3736-1	KP876506, KU5	Alphacoronavirus	1	Ge et al. (2016)	WIV	3736	1	CN	YN	TG	Tongguan	Yunnan	Southern	06-Aug-12	Aug-12
Miniopterus bat coronavirus BtCoV/3736-2	KP876505	Alphacoronavirus	HKU8	Ge et al. (2016)	WIV	3736	2	CN	YN	TG	Tongguan	Yunnan	Southern	06-Aug-12	Aug-12
Hipposideros bat coronavirus BtCoV/3740-1	KP876530, KU5	Alphacoronavirus	HKU10	Ge et al. (2016)	WIV	3740	1	CN	YN	TG	Tongguan	Yunnan	Southern	06-Aug-12	Aug-12
Hipposideros bat coronavirus BtCoV/3740-2	KP876531, KU5	Betacoronavirus	Lineag	Ge et al. (2016)	WIV	3740	2	CN	YN	TG	Tongguan	Yunnan	Southern	06-Aug-12	Aug-12
Rhinolophus bat coronavirus BtCoV/3750	KP876539, KU5	Alphacoronavirus	HKU2	Ge et al. (2016)	WIV	3750		CN	YN	TG	Tongguan	Yunnan	Southern	06-Aug-12	Aug-12
Rhinolophus bat coronavirus HKU2 3755	MF094702	Alphacoronavirus	SADSr	Zhou et al. (2018), Wang	WIV+EcoH+DNUS	3755		CN	YN	TG	Tongguan	Yunnan	Southern	06-Aug-12	Aug-12
Miniopterus bat coronavirus BtCoV/3759-1	KP876514, KU5	Alphacoronavirus	1	Ge et al. (2016)	WIV	3759	1	CN	YN	TG	Tongguan	Yunnan	Southern	06-Aug-12	Aug-12
Miniopterus bat coronavirus BtCoV/3759-2	KP876515	Alphacoronavirus	HKU8	Ge et al. (2016)	WIV	3759	2	CN	YN	TG	Tongguan	Yunnan	Southern	06-Aug-12	Aug-12
Miniopterus bat coronavirus BtCoV/3760-1	KP876512	Alphacoronavirus	1	Ge et al. (2016)	WIV	3760	1	CN	YN	TG	Tongguan	Yunnan	Southern	06-Aug-12	Aug-12
Miniopterus bat coronavirus BtCoV/3760-2	KP876513	Alphacoronavirus	HKU8	Ge et al. (2016)	WIV	3760	2	CN	YN	TG	Tongguan	Yunnan	Southern	06-Aug-12	Aug-12
Miniopterus bat coronavirus BtCoV/3767	KP876542	Alphacoronavirus	HKU8	Ge et al. (2016)	WIV	3767		CN	YN	TG	Tongguan	Yunnan	Southern	06-Aug-12	Aug-12
Rhinolophus bat coronavirus BtCoV/3772	KP876540	Alphacoronavirus	HKU2	Ge et al. (2016)	WIV	3772		CN	YN	TG	Tongguan	Yunnan	Southern	06-Aug-12	Aug-12
Coronavirus PREDICT CoV-56 PREDICT_CoV-56/LAP12-G1-0029	KX286290	Betacoronavirus		Anthony et al. (2017)	PREDICT			LA				_Laos_		07-Aug-12	Aug-12
Paramyxovirus PREDICT_PMV-31 isolate PREDICT_PMV-31/EHA-15(	KP963807	Paramyxovirus					1	CN	YN	FG	Fugong	Yunnan	Western	09-Aug-12	Aug-12
Hantavirus China/YN06_Hantavirus/PREDICT_EHA-156-12-YN-FG0	KX442788	Orthohantavirus		Ge et al. (2016b)	WIV+EcoH		1	CN	YN	FG	Fugong	Yunnan	Western	09-Aug-12	Aug-12
Vole astrovirus Ec/FG004/Yunnan	KJ571436, KY(	Astrovirus		Hu et al. (2014)	WIV+EcoH		4	CN	YN	FG	Fugong	Yunnan	Western	09-Aug-12	Aug-12
Vole astrovirus Ec/FG005/Yunnan	KJ571437, KY(	Astrovirus		Hu et al. (2014)	WIV+EcoH		5	CN	YN	FG	Fugong	Yunnan	Western	09-Aug-12	Aug-12
Hantavirus China/YN06_Hantavirus/PREDICT_EHA-156-12-YN-FG0	KX442784	Orthohantavirus		Ge et al. (2016b)	WIV+EcoH		5	CN	YN	FG	Fugong	Yunnan	Western	09-Aug-12	Aug-12
Vole astrovirus Ec/FG006/Yunnan	KJ571438, KY(	Astrovirus		Hu et al. (2014)	WIV+EcoH		6	CN	YN	FG	Fugong	Yunnan	Western	09-Aug-12	Aug-12

> What if those SARS2-like sequences were not actually in the pangolins and were just mere lab cotamination of a close relative to SARS2 used in the lab? (we have recently seen many other cases of contamination in other samples)
> GIABR have been sampling in Yunnan, even in Mojiang pic.twitter.com/sErzG5ZQVH
>
> — Francisco de Asis (@franciscodeasis) November 27, 2020

LiBiao Zhang was also sampling in the same location as Holmes. GPS coordinates are 4 km apart, although it seems that Holmes' team obfuscated a bit the location. He may even have sampled a close relative bat unnoticing that was infected with similar virus
https://t.co/KDNirmZ9DX

> Pangolin CoV from lab contamination is increasingly evident:
> Liang et al. (2019) [Libiao Zhang] collected bats in Apr-15 in the same location as Zhou et al. (2020b) [Holmes] collected RmYN02 in 2019, with a 100% ident. cyt bhttps://t.co/CQuAbKcUAuhttps://t.co/qZaTEidKZ1
> pic.twitter.com/TEej6YQAmz
>
> — Francisco de Asis (@franciscodeasis) February 6, 2021

Also, Libiao Zhang was in Yunnan in July 2019 catching bats with students. So, we have not only two possible sources of lab contamination, but also we cannot rule out a lab zoonosis if any bats were sent to GIABR/GIZ and kept in the same room as pangolins
https://t.co/i7ZdsTURKi



Thanks @BillyBostickson for the link

Another possibility is that one of these students (Libiao Zhang is probably already well immunized) served as a human intermediate host for the pangolins. Not being able to infect h2h but h2p? I think it is less likely but cannot rule out either
https://t.co/AvHO3pe6lA

> About shoddy safety protocols:

> Clumsy students and researchers from Wuhan collecting bat samples with inadequate PPE (sometime around June-July 2019)https://t.co/FsK7jw11hI
>
> — The Seeker (@TheSeeker268) May 18, 2020

Also pointed by @TheSeeker: "the binding affinity of the pangolin ACE2 receptor for SARS-CoV-2 RBD was later on reported to be low" https://t.co/d0yMNAqZeG

There is a fifth possible source of the contamination, but from a Bayesian point of view it should be less likely. We will explain it in an special thread on lab contamination soon

I forgot to add this to this threat: https://t.co/aZzyhLMGmK

> Oops, what a coincidence! Libiao Zhang (from GIABR, the lab of pangolins) forgot to mention his paper with WIV in which they isolated viruses collected from "Mojiang" (sic). He did not forget the other paper with them (in same journal a few issues before) https://t.co/iAPXAmMamk
>
> — Francisco de Asis (@franciscodeasis) January 25, 2021

Just found a great coincidence, although it could be spurious: Jin-Ping Chen is corresponding author of Yuan et al. (2014) with ZLS and Daszak. He was nearby while the miners outbreak was taking place. Although, they aparently did not sampled Ra, Rs or Rm https://t.co/HC52X3sbgP

> So, it is possible that WIV & EcoH stopped in TG between 07-Apr-12 & 11-Apr-12 when moving from Yuanjiang to Mengla. In those days the miners started "re-cleaning" the mineshaft. I guess that "1st day in mine" & "1st symptoms" dates are approx.https://t.co/Ea9rqNzZYX
>
> — Francisco de Asis (@franciscodeasis) September 3, 2020

To end, something very important: Do not discard this "Pangolin CoV" story! The metagenomes are probably the most trustworthy data along with 4991 RdRp.
Uploaded on 2019-09-23!

COVID-19 is an emerging, rapidly evolving situation.

Public health information (CDC) | Research information (NIH) | SARS-CoV-2 data (NCBI) |

Virome of dead pangolin individuals Metagenome

Identifiers:	SRA:	SRP223042
	BioProject:	PRJNA573298
Study Type:	Other	
Abstract:	Viral diversity and pathogens of dead Malay pangolin samples	
External Link:	Viral Metagenomics Revealed Sendai Virus and Coronavirus Infection of *javanica*).	

Related Files

Organism	Sample	File Name	Size	Updated
Manis javanica	SAMN12809949	lung02.1.fq.gz	2.5 Gb	2019-09-23 22:46:44
Manis javanica	SAMN12809949	lung02.2.fq.gz	2.5 Gb	2019-09-23 22:47:16
Manis javanica	SAMN12809952	lung07.1.fq.gz	911.6 **Mb**	2019-09-23 22:32:36
Manis javanica	SAMN12809952	lung07.2.fq.gz	935.8 **Mb**	2019-09-23 22:32:18
Manis javanica	SAMN12809953	lung08.1.fq.gz	768.6 **Mb**	2019-09-23 22:31:02
Manis javanica	SAMN12809953	lung08.2.fq.gz	780.6 **Mb**	2019-09-23 22:31:05
Manis javanica	SAMN12809954	lung09.1.fq.gz	848.7 **Mb**	2019-09-23 22:33:06
Manis javanica	SAMN12809954	lung09.2.fq.gz	872.4 **Mb**	2019-09-23 22:33:23
Manis javanica	SAMN12809955	lung11.1.fq.gz	1.1 Gb	2019-09-23 22:31:07
Manis javanica	SAMN12809955	lung11.2.fq.gz	1.2 Gb	2019-09-23 22:31:30