Prevalence of *mcr-1* in Colonized Inpatients, China, 2011–2019

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In response to the spread of colistin resistance gene *mcr-1*, China banned the use of colistin in livestock fodders. We used a time-series analysis of inpatient colonization data from 2011–2019 to accurately reveal the associated fluctuations of *mcr-1* that occurred in inpatients in response to the ban.

Heavy use of antimicrobials in agricultural, human, and veterinary applications correlates directly with emergence and spread of antimicrobial resistance, thereby threatening the effective management of clinical infections (*1*,*2*). An example of this association is the global dissemination of the antimicrobial resistance gene (ARG) *mcr-1*, conferring resistance to the last-line antimicrobial drug colistin. The *mcr-1* gene has been prevalent in ecosystems that use colistin as a growth promoter in food-producing animals, as seen in China before 2017 (*2*–*5*).

To counteract the high prevalence of *mcr-1* and align with One Health principles, the government in China formally banned colistin as an animal feed additive on April 30, 2017 (*6*). Previous research demonstrated that colistin resistance rates and *mcr-1* prevalence in *Escherichia coli* from human and animal samples declined substantially in China, according to a regional study conducted in Guangzhou during 2015–2019 ($p<0.0001$). These data suggest the effectiveness of colistin stewardship in reducing colistin resistance in both livestock and humans (*4*,*5*). However, the sampling strategy of these studies was limited to evaluating only several cross-sectional timepoints from before and after the ban, resulting in uncertainty about the exact timing of the effect.

To characterize the complete prevalence dynamics of human *mcr-1* colonization, including the periban period, we constructed a 9-year monthly time series for April 2011–December 2019, over which time 13,630 fecal samples from colonized inpatients were previously taken, by further evaluating *mcr-1* prevalence of 3,823 stored fecal samples collected during April–September 2016, January–September 2017–2018, and January–December 2019. We combined these data with those from our previous studies (*3*,*5*) (Appendix Table 1, https://wwwnc.cdc.gov/EID/article/27/9/20-3642-App1.pdf). We used a 3-month moving average approach to remove noise and substituted missing data for 7 months of the time series by using the mean values of the 2 months flanking any month with missing data (Appendix). Through changepoint analysis (Appendix) (*7*), we identified 5 changepoints, dividing the time series into 6 periods (Figure).

We observed that *mcr-1* prevalence in human fecal samples was low (<3%) in the early period, before October 2013, demonstrating that the *mcr-1* gene was circulating to a limited extent in human populations before late 2013 in period 1 (P1). We observed a significant increase in *mcr-1* colonization prevalence after November 2013 in period 2 (P2) that lagged behind increases of *mcr-1* prevalence observed in livestock from 2011 (*2*) and was consistent with dissemination from this reservoir. The third period (P3) showed a sharp increase in *mcr-1* human colonization prevalence, followed by a peak in October 2016, suggesting that *mcr-1* was rapidly spreading in human settings, potentially attributable to an extremely high *mcr-1* prevalence (>60%) in livestock around the time (*4*,*5*,*8*). Beginning in November 2016, in period 4 (P4), pilot decreases in colistin use as an animal feed additive were already being implemented (*4*) before the complete ban in 2017. We observed declines in human *mcr-1* colonization prevalence during this period

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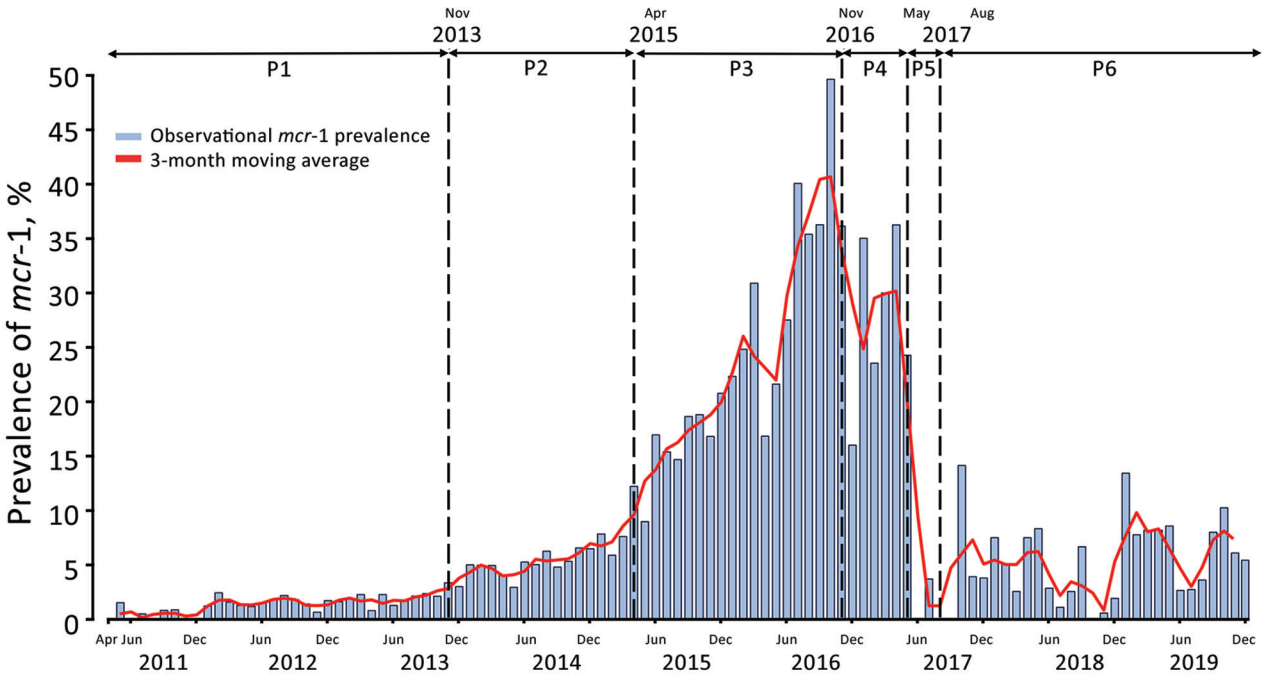

Figure. Time series of monthly *mcr-1* prevalence in colonized inpatients, China, April 2011–December 2019. The *mcr-1* prevalence was recorded each month in observed data (blue histogram) and 3-month moving average data (solid red line). Vertical dashed lines indicate significant changepoints identified in the changepoint analysis: November 2013, May 2015, November 2016, May 2017, and August 2018. The government of China formally banned colistin as an animal feed additive on April 30, 2017. P, time period.

that were temporally consistent with declines in *mcr-1* prevalence observed in livestock (*8*). The fifth period (P5) showed a dramatic decline in human *mcr-1* colonization prevalence, correlating with the complete ban of colistin in animal feed (*6*). The rapid impact of this intervention is indicative of the dramatic effect that curtailing a selection pressure can have in constraining ARG prevalence and could be a template for combatting other ARGs. In the last period evaluated, period 6 (P6), *mcr-1* prevalence fluctuated at a low level (monthly average 5.3%), in accordance with the *mcr-1* prevalence observed in healthy human carriers, pigs, and chickens after the colistin ban (*5*). Alhough currently at low levels, *mcr-1* prevalence should be monitored continually to detect any signs of its resurgence, particularly given that colistin was approved for human clinical use in China in January 2017 (*9*).

In conclusion, we characterized the dynamic landscape of *mcr-1* over a 9-year period in China and found that colistin stewardship interventions in livestock were reflected in the *mcr-1* prevalence in human fecal colonization samples within a month of a large-scale, national ban on colistin usage. Partial reductions in colistin use beginning in November 2016 rapidly reduced the *mcr-1* prevalence and turned around the alarming increases observed during 2015–2016. The complete ban implemented on April 30, 2017, significantly and immediately reduced *mcr-1* prevalence to near pre-2015 levels. Of interest, however, the background *mcr-1* prevalence in 2019 was still higher than that observed during 2011–2013, perhaps associated with the approval of colistin for human clinical use in China in January 2017 (*9*). As a result of our findings, we strongly encourage interdisciplinary surveillance involving clinicians, veterinary specialists, and environmentalists to further investigate and evaluate changes in ARG prevalence across different human, animal, and environmental niches to improve holistic understanding of the impact and timeframe of different stewardship interventions.

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References

1. Tang KL, Caffrey NP, Nóbrega DB, Cork SC, Ronksley PE, Barkema HW, et al. Restricting the use of antibiotics in food-producing animals and its associations with antibiotic resistance in food-producing animals and human beings: a systematic review and meta-analysis. Lancet Planet Health. 2017;1:e316–27. https://doi.org/10.1016/S2542-5196(17)30141-9
2. Shen Z, Wang Y, Shen Y, Shen J, Wu C. Early emergence of *mcr-1* in *Escherichia coli* from food-producing animals. Lancet Infect Dis. 2016;16:293. https://doi.org/10.1016/S1473-3099(16)00061-X
3. Zhong LL, Phan HTT, Shen C, Vihta KD, Sheppard AE, Huang X, et al. High rates of human fecal carriage of *mcr-1*-positive multidrug-resistant *Enterobacteriaceae* emerge in China in association with successful plasmid families. Clin Infect Dis. 2018;66:676–85. https://doi.org/10.1093/cid/cix885
4. Wang Y, Xu C, Zhang R, Chen Y, Shen Y, Hu F, et al. Changes in colistin resistance and *mcr-1* abundance in *Escherichia coli* of animal and human origins following the ban of colistin-positive additives in China: an epidemiological comparative study. Lancet Infect Dis. 2020;20:1161–71. https://doi.org/10.1016/S1473-3099(20)30149-3
5. Shen C, Zhong L, Yang Y, Doi Y, Paterson D, Stoesser N, et al. Dynamics of *mcr-1* prevalence and *mcr-1*-positive *Escherichia coli* after the cessation of colistin use as a feed additive for animals in China: a prospective cross-sectional and whole genome sequencing-based molecular epidemiological study. Lancet Microbe. 2020;1:e34–43. https://doi.org/10.1016/S2666-5247(20)30005-7
6. Walsh TR, Wu Y. China bans colistin as a feed additive for animals. Lancet Infect Dis. 2016;16:1102–3. https://doi.org/10.1016/S1473-3099(16)30329-2
7. Killick R, Eckley I. Changepoint: an R package for changepoint analysis. J Stat Softw. 2014;58:1–19. https://doi.org/10.18637/jss.v058.i03
8. Li W, Hou M, Liu C, Xiong W, Zeng Z. Dramatic decrease in colistin resistance in *Escherichia coli* from a typical pig farm following restriction of colistin use in China. Int J Antimicrob Agents. 2019;53:707–8. https://doi.org/10.1016/j.ijantimicag.2019.03.021
9. Zhang R, Shen Y, Walsh TR, Wang Y, Hu F. Use of polymyxins in Chinese hospitals. Lancet Infect Dis. 2020;20:1125–6. https://doi.org/10.1016/S1473-3099(20)30696-4

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Haemophilus influenzae Type a Sequence Type 23, Northern Spain

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Two consecutive cases of *Haemophilus influenzae* type a sequence type 23 invasive infection in 2 children attending the same daycare in 2019 triggered epidemiologic surveillance of *H. influenzae* infections in northern Spain. Despite the invasiveness potential of this virus strain, we detected no additional cases for 2013–2020.

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Since the introduction of the *Haemophilus influenzae* type b (Hib) conjugate vaccine in the infant immunization schedule in 1998, the incidence of invasive *H. influenzae* (Hi) infections in Spain decreased to 0.7 episodes/100,000 population (*1*). Higher incidence rates are observed in children ≤2 years of age (1.88/100,000 population) and adults ≥65 years of age (1.89 cases/100,000 population) (*2*). Invasive disease caused by Hib has nearly disappeared, and most cases are caused by nontypeable strains (*3*).

Invasive infections caused by *H. influenzae* type a (Hia) are uncommon in Europe, particularly in Spain. However, Hia incidence is as high in other regions as among indigenous communities in North America (*4*) and as has emerged in Brazil during the 2000s (*5*). We describe 2 cases of Hia invasive disease in Gipuzkoa, northern Spain.

Both cases of Hia invasive disease occurred in children in a village with ≈15,000 inhabitants during November 2–3, 2019. The first patient, a 2-year-old boy, was admitted to the pediatric emergency department with good general aspect and persistent low-grade fever without a clear source. The child was not vaccinated according to the routine immunization schedule. Results for pulmonary auscultation and respiratory and cardiac rates were unremarkable, and a chest radiograph showed no abnormalities.

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Appendix

Data collection

To investigate the longitudinal dynamics of *mcr-1* prevalence in *mcr-1*-colonized patients, we retrospectively analyzed 13,630 samples from April 2011 to December 2019 monthly in Guangzhou, China, except for the months of January 2012, February 2013, February 2014, March 2017, February 2018, and March and April in 2019, for which samples/data were not available. Prevalence data from April 2011 to March 2016 (except January 2012, February 2013, and February 2014 as above), were acquired from our previous study (*1*), in which 8,022 human fecal samples were obtained from three tertiary hospitals in Guangzhou, China. Among them, 497 samples were identified as *mcr-1*-positive.

The prevalence data from October to December in 2016–2018 were obtained from another previous study (*2*), in which we identified 364/2395 *mcr-1*-positive human fecal samples from two hospitals which were also sites in the study cited above (*1*).

mcr-1 prevalence data from April to September in 2016–2018 and from January to December in 2019 were obtained additionally for this study (except for data from February 2018, March and April in 2019, missing - as above). We collected 3823 samples from human colonization samples in two hospitals. The sampling hospitals and the inclusion criteria are consistent with above study (*2*).

Sample collection, sample culture, *mcr-1* screening for the samples in this study

Sampling procedures and *mcr-1* identification were consistent with our previous studies (*1*,*2*). Briefly, human fecal samples were collected using rectal swabs from inpatients without any diagnosed infections. All samples were cultured on non-selective nutrient broth media overnight at 37°C. Total DNA was extracted using the boiling method and screened for *mcr-1* by polymerase chain reaction (PCR) as previously described.[2] *mcr-1*-positive samples were re-cultured on selective agar (MacConkey + colistin [2 mg/L]) and incubated for 18–24 hours, 37°C). Subsequently, up to 10 Enterobacteriaceae colonies were sub-cultured to MacConkey agar with colistin (2 mg/L). The samples were identified as positive for *mcr-1* if any one colony was positive for *mcr-1* by PCR.

Time series analysis

Missing data for Jan 2012, February 2013, February 2014, March 2017 and February 2018 were substituted by the mean values of the two relevant flanking months. The missing data of March and April in 2019 were substituted by the mean values of February and May in 2019. A 3-month moving average (MA) approach was used to remove noise.

We conducted a changepoint detection analysis to identify the time points of significant changes in the monthly overall *mcr-1* prevalence from April 2011 to December 2019. For this analysis, we used the *cpt.meanvar* function from the changepoint package and explored a variety of penalty values and methods (*3*), retaining the most consistently identified changepoints with the proposed pruned exact linear time (PELT) algorithm. As a result, we identified five changepoints in the time series, which was divided into six periods [period 1 (April 2011 to November 2013), period 2 (December 2013 to April 2015), period 3 (May 2015 to November 2016), period 4 (December 2016 to May 2017), period 5 (June 2017 to August 2017), period 6 (September 2017 to December 2019)] (Figure 1).

Appendix Table. Data underpinning the combined *mcr-1* prevalence monthly time series in human fecal colonization samples from Guangzhou, China, April 2011-December 2019.

Period	Date	Sample size	*mcr-1*-positive sample	*mcr-1* prevalence % (95%CI)	Three-month moving average %	Data reference
1	Apr 2011	138	0	0	/	Zhong LL, et al. Clin Infect Dis. 2018; 66: 676–85.
	May 2011	66	1	1.52 (0–4.46)	0.51	
	Jun 2011	144	0	0	0.67	
	Jul 2011	201	1	0.5 (0–1.47)	0.17	
	Aug 2011	183	0	0	0.44	
	Sep 2011	122	1	0.82 (0–2.42)	0.56	
	Oct 2011	232	2	0.86 (0–2.05)	0.56	
	Nov 2011	168	0	0	0.29	
	Dec 2011	39	0	0	0.41	
	Jan 2012	N.D.	N.D.	1.22	1.22	
	Feb 2012	82	2	2.44 (0–5.78)	1.75	
	Mar 2012	63	1	1.59 (0–4.67)	1.77	
	Apr 2012	156	2	1.28 (0–3.05)	1.35	
	May 2012	85	1	1.18 (0–3.47)	1.31	
	Jun 2012	202	3	1.49 (0–3.15)	1.49	
	Jul 2012	166	3	1.81 (0–3.83)	1.83	
	Aug 2012	183	4	2.19 (0.07–4.3)	1.93	
	Sep 2012	102	0	1.8	1.80	
	Oct 2012	213	3	1.41 (0–2.99)	1.28	
	Nov 2012	155	1	0.65 (0–1.91)	1.25	
	Dec 2012	117	2	1.71 (0–4.06)	1.32	
	Jan 2013	62	1	1.61 (0–4.75)	1.76	
	Feb 2013	N.D.	N.D.	1.94	1.94	
	Mar 2013	88	2	2.27 (0–5.39)	1.67	
	Apr 2013	125	1	0.8 (0–2.36)	1.78	
	May 2013	88	2	2.27 (0–5.39)	1.45	
	Jun 2013	156	2	1.28 (0–3.05)	1.74	
	Jul 2013	181	3	1.66 (0–3.52)	1.69	
	Aug 2013	141	3	2.13 (0–4.51)	2.05	
	Sep 2013	127	3	2.36 (0–5)	2.20	
	Oct 2013	189	4	2.12 (0.06–4.17)	2.61	
2	Nov 2013	209	7	3.35 (0.91–5.79)	2.83	Zhong LL, et al. Clin Infect Dis. 2018; 66: 676–85.
	Dec 2013	166	5	3.01 (0.41–5.61)	3.79	

Period	Date	Sample size	*mcr-1*-positive sample	*mcr-1* prevalence % (95%CI)	Three-month moving average %	Data reference
	Jan 2014	80	4	5 (0.22–9.78)	4.33	
	Feb 2014	N.D.	N.D.	4.97	4.97	
	Mar 2014	182	9	4.95 (1.79–8.09)	4.67	
	Apr 2014	147	6	4.08 (0.88–7.28)	3.99	
	May 2014	102	3	2.94 (0–6.22)	4.10	
	Jun 2014	133	7	5.26 (1.47–9.06)	4.41	
	Jul 2014	159	8	5.03 (1.63–8.43)	5.51	
	Aug 2014	128	8	6.25 (2.06–10.4)	5.36	
	Sep 2014	167	8	4.79 (1.55–8.03)	5.45	
	Oct 2014	188	10	5.32 (2.11–8.53)	5.55	
	Nov 2014	168	11	6.55 (2.81–10.9)	6.12	
	Dec 2014	108	7	6.48 (1.84–11.12)	6.96	
	Jan 2015	102	8	7.84 (2.63–13.06)	6.74	
	Feb 2015	68	4	5.88 (0.29–11.47)	7.11	
	Mar 2015	79	6	7.59 (1.75–13.44)	8.56	
	Apr 2015	131	16	12.21 (6.61–17.82)	9.59	
3	May 2015	156	14	8.97 (4.49–13.46)	12.71	Zhong LL, et al. Clin
	Jun 2015	118	20	16.95 (10.18–23.72)	13.77	Infect Dis. 2018; 66:
	Jul 2015	117	18	15.38 (8.85–21.92)	15.67	676–85.
	Aug 2015	177	26	14.69 (9.47–19.9)	16.24	
	Sep 2015	161	30	18.63 (12.62–24.65)	17.38	
	Oct 2015	218	41	18.81 (13.62–23.99)	18.08	
	Nov 2015	238	40	16.81 (12.06–21.56)	18.79	
	Dec 2015	183	38	20.77 (14.89–26.64)	19.97	
	Jan 2016	103	23	22.33 (14.29–30.37)	22.64	
	Feb 2016	137	34	24.82 (17.58–32.05)	26.01	
	Mar 2016	123	38	30.89 (22.73–39.06)	24.18	
	Apr 2016	297	50	16.84 (12.58–21.09)	23.11	This study
	May 2016	236	51	21.61 (16.36–26.86)	21.98	
	Jun 2016	349	96	27.51 (22.82–32.19)	29.73	
	Jul 2016	327	131	40.06 (34.75–45.37)	34.32	
	Aug 2016	407	144	35.38 (30.74–40.03)	37.23	
	Sep 2016	262	95	36.26 (30.44–42.08)	40.43	
	Oct 2016	278	138	49.64 (43.76-55.52)	40.68	Shen C, et al. The Lancet Microbe. 2020;1: e34–43.
4	Nov 2016	404	149	36.14 (31.1-41.18)	33.93	

Period	Date	Sample size	*mcr-1*-positive sample	*mcr-1* prevalence % (95%CI)	Three-month moving average %	Data reference
	Dec 2016	99	16	16.16 (8.91-23.41)	29.05	Shen C, et al. The Lancet Microbe. 2020;1: e34–43.
	Jan 2017	78	27	34.62 (24.06-45.18)	24.85	This study
	Feb 2017	68	17	25 (14.71-35.29)	29.52	
	Mar 2017	N.D.	N.D.	30.63	29.93	
	Apr 2017	80	29	36.25 (25.72-46.78)	30.18	
5	May 2017	173	42	24.28 (17.89–30.67)	20.18	This study
	Jun 2017	44	0	0	9.33	
	Jul 2017	54	2	3.7 (0–8.74)	1.23	
6	Aug 2017	57	0	0	1.23	This study
	Sep 2017	20	0	0	4.72	
	Oct 2017	106	15	14.15 (7.51-20.79)	6.02	Shen C, et al. The Lancet Microbe. 2020;1: e34–43.
	Nov 2017	277	11	3.92 (1.63-6.21)	7.29	
	Dec 2017	246	9	3.66 (1.6–6.53)	5.07	
	Jan 2018	40	3	7.5 (0–15.66)	5.44	This study
	Feb 2018	N.D.	N.D.	5.03	5.03	
	Mar 2018	78	2	2.56 (0–6.07)	5.03	
	Apr 2018	40	3	7.5 (0–15.66)	6.13	
	May 2018	60	5	8.33 (1.34–15.33)	6.23	
	Jun 2018	70	2	2.86 (0–6.76)	4.10	
	Jul 2018	90	1	1.11 (0–3.28)	2.18	
	Aug 2018	39	1	2.56 (0–7.52)	3.45	
	Sep 2018	90	6	6.67 (1.51–11.82)	3.08	
	Aug 2018	62	0	0	2.41	Shen C, et al. The Lancet Microbe. 2020;1: e34–43.
	Nov 2018	175	1	0.57 (0–1.69)	0.83	
	Dec 2018	138	3	2.17 (0–4.6)	5.31	
	Jan 2019	67	9	13.43 (5.27–21.6)	7.71	This study
	Feb 2019	90	7	7.78 (2.24–13.31)	9.8	
	Mar 2019	N.D.	N.D.	8.17	8.04	
	Apr 2019	N.D.	N.D.	8.17	8.31	
	May 2019	70	6	8.57 (2.01–15.13)	6.47	
	Jun 2019	113	3	2.65 (0–5.62)	4.66	
	Jul 2019	73	2	2.74 (0–6.48)	3	
	Aug 2019	83	3	3.61 (0–7.63)	4.78	
	Sep 2019	25	2	8 (0–18.63)	7.29	
	Oct 2019	39	4	10.26 (0.73–19.78)	8.12	

Period	Date	Sample size	*mcr-1*-positive sample	*mcr-1* prevalence % (95%CI)	Three-month moving average %	Data reference
	Nov 2019	82	5	6.1 (0.92–11.28)	7.25	
	Dec 2019	222	12	5.41 (2.43–8.38)	/	

References

1. Zhong LL, Phan HTT, Shen C, Vihta KD, Sheppard AE, Huang X, et al. High rates of human fecal carriage of *mcr-1*-positive multidrug-resistant Enterobacteriaceae emerge in China in association with successful plasmid families. Clin Infect Dis. 2018;66:676–85. PubMed https://doi.org/10.1093/cid/cix885

2. Shen C, Zhong LL, Yang Y, Doi Y, Paterson D, Stoesser N, et al. Dynamics of *mcr-1* prevalence and *mcr-1*-positive *Escherichia coli* after the cessation of colistin use as a feed additive for animals in China: a prospective cross-sectional and whole genome sequencing-based molecular epidemiological study. Lancet Microbe. 2020;1:e34–43. https://doi.org/10.1016/S2666-5247(20)30005-7

3. Killick R, Eckley I. Changepoint: an R package for changepoint analysis. J Stat Softw. 2014;58:1–19. https://doi.org/10.18637/jss.v058.i03