International Journal of Antimicrobial Agents 57 (2021) 106167

Contents lists available at ScienceDirect

International Journal of Antimicrobial Agents

journal homepage: www.elsevier.com/locate/ijantimicag

Letter to the Editor

Advanced statistical methods and designs for clinical trials for COVID-19

Hydroxychloroquine alone and a combination of hydroxychloroquine and azithromycin were shown to be more effective than the control in treating patients with COVID-19 virus in a small sample non-randomized study reported by Gautret et al. [1]. Hydroxychloroquine, approved by the Food and Drug Administration (FDA) as an anti-malarial drug, was repurposed to treat patients with COVID-19 virus. This important finding along with results from other treatments (e.g., Chloroquine) may point out a path to end the coronavirus pandemic. However, we find that the statistical analyses by Gautret et al. [1] are inappropriate. One of the three goals of this letter is to fix the errors so that their promising clinical results are properly interpreted. A follow-up large-scale clinical trial has started in the United States to confirm its activity [2]. For an early phase clinical trial, proper and efficient statistical methods would provide valid results and novel study designs to speed up the treatment discovery.

The proportions of patients with PCR-negative were calculated for each group: 16 patients in the control group, 14 patients treated by hydroxychloroquine only, and 6 patients treated by a combination of hydroxychloroquine and azithromycin. The proportions were compared by using the chi-squared test with p-values in Table 2 in the article by Gautret et al. [1]. There are three concerns. First, their p-values were computed for a two-sided alternative hypothesis. i.e., a small p-value, e.g., p=0.04 for Day 4, only means that the hydroxychloroquine treated patients have a different PCR-negative rate from the control patients. However, we wish to see the former has a larger rate, which is supported by the p-values for the one-sided alternative hypothesis: the hydroxychloroquine treated patients have a larger PCR-negative rate than the control patients. Secondly, the validity of the chi-squared test requires large sample sizes, which did not occur in the study [1]. For a study with small sample sizes, exact tests (e.g., Barnard test), are needed to control the type I error. Lastly, statistical approaches should be aligned with the study design. The hydroxychloroquine trial was designed with the sample sizes fixed in each group. The Barnard test is appropriate here as it is unconditional and only assumes the fixed sample sizes of each group, while the traditionally used Fisher test is an exact conditional test, which assumes both the sample sizes and the total number of responses are fixed. For these reasons, we would recommend using the Barnard exact unconditional test for the p-value calculation and the results are presented in Table 1.

Confidence intervals for the difference of two proportions contain more information than statistical tests for testing the effectiveness of hydroxychloroquine to treat COVID-19 patients. Due to small sample sizes, it is recommended to use the exact confidence interval by Wang [3] for the difference. In the hydroxychloroquine trial [1], since hydroxychloroquine was expected to increase the PCR-negative rate as compared to the control, a lower one-sided confidence limit would be proper to be reported. e.g., the Wang exact 95% one-sided lower limit for the proportion difference between the hydroxychloroquine group and the control group at Day 6, is computed as [0.3137,1], see Table 1. It is calculated by using the statistical software R package ExactCIdiff [4]. The interval is positive, so it confirms the effectiveness of hydroxychloroquine as the Barnard test does. In addition, the interval also concludes that the use of hydroxychloroquine increases the PCR-negative rate by at least 31.37% with 95% confidence. Due to the small sample size in the study, the exact one-sided confidence limits are recommended for use with valid statistical inference.

In addition to age, blood type is another risk factor in COVID-19: people with blood A type have a much higher risk to catch COVID-19 virus than people with non-A type (odds ratio of 1.2 in average) [5]. In the hydroxychloroquine trial, three out of the six patients dropped the study were transferred to intensive care unit. Disease severity may be another important factor that needs to be considered in the future study designs. When a new treatment only benefits subpopulations of COVID-19 patients stratified by the known risk factors (e.g., age and blood type), adaptive enrichment designs could be utilized to quickly identify these subpopulations, and assign the remaining subpopulations to other potentially effective treatments.

In conclusion, we discussed the proper statistical methods (the Barnard test and the Wang interval) for clinical trials with small sample sizes to increase the success rate of trials with valid statistical inference. Since COVID-19 is a severe public safety issue, it is very important to approve or disapprove clinical results through solid statistical procedures. In addition, new and novel clinical designs (e.g., adaptive designs) are encouraged to speed up the discovery of new treatments for COVID-19 virus and others.

DOI of original article: 10.1016/j.ijantimicag.2020.105949

https://doi.org/10.1016/j.ijantimicag.2020.106167
0924-8579/© 2020 Elsevier Ltd and International Society of Antimicrobial Chemotherapy. All rights reserved.

Table 1
Proportion differences of PCR negative patients in the hydroxychloroquine group and the control group, reported two-sided p-values by Gautret et al. [1], Barnard exact one-sided p-values for proportion difference, and Wang exact one-sided confidence intervals for proportion difference.

	Day 3	Day 4	Day 5	Day 6
Proportion difference	43.75%	45.00%	46.25%	57.50%
Reported two-sided p-value	0.0046	0.0357	0.0055	0.0006
Barnard one-sided p-value	0.0025	0.0218	0.0031	0.0002
Wang one-sided confidence interval	[0.1812,1]	[0.0700,1]	[0.1905,1]	[0.3137,1]

References

[1] Gautret P, Lagier JC, Parola P, Hoang V, Meddeb L, Mailhe M, et al. Hydroxychloroquine and azithromycin as a treatment of COVID-19: results of an open-label non-randomized clinical trial. Int J Antimicrob Agents 2020;56(1):105949.
[2] Harding R. Cuomo: NY to conduct drug trials for coronavirus treatment. (Mar 22, 2020) https://auburnpub.com/news/local/govt-and-politics/cuomo-ny-to-conduct-drug-trials-for-coronavirus-treatment/article_0fb6c870-7e29-5ee5-8bda-bbda132a5418.html.
[3] Wang W. On construction of the smallest one-sided confidence interval for the difference of two proportions. The Annals of Statistics 2010;38:1227–43.
[4] Shan G, Wang W. ExactCIdiff: an R package for computing exact confidence intervals for the difference of two proportions. The R Journal 2013;5(2):62–71.
[5] Zhao J, Yang Y, Huang HP, Li D, Gu DF, Lu XF, Zhang Z, Liu L, Liu T, Liu YK, He YJ. Relationship between the ABO Blood Group and the COVID-19 Susceptibility. medRxiv. January 2020.

Guogen Shan*
Epidemiology and Biostatistics Program, Department of Environmental and Occupational Health, School of Public Health, University of Nevada Las Vegas, Las Vegas, 89154, NV, USA

Weizhen Wang
Department of Mathematics and Statistics, Wright State University, Dayton, OH 45435, USA

*Corresponding author.
E-mail addresses: guogen.shan@unlv.edu (G. Shan), weizhen.wang@wright.edu (W. Wang)