

EVALUATION OF ANTIBIOTIC RESISTANCE CASES IN ACUTE RESPIRATORY INFECTION DISEASE: POTENTIAL TEST AND COMPARATIVE TEST OF TETRACYCLINE ANTIBIOTIC

Danni Ramdhani*¹, Sri Agung F. K.² and Ika Khumairoh²

¹Department of Pharmaceutical Analysis and Medicinal Chemistry, Faculty of Pharmacy.

²Departement of Pharmaceutical Biology, Faculty of Pharmacy, Universitas Padjadjaran, Sumedang, Indonesia 45363.

Article Received on
11 Dec. 2018,

Revised on 31 Dec. 2018,
Accepted on 21 Jan. 2019

DOI: 10.20959/wjpr20192-14174

*Corresponding Author

Dr. Danni Ramdhani

Department of
Pharmaceutical Analysis and
Medicinal Chemistry,
Faculty of Pharmacy.

ABSTRACT

Objective: Acute respiratory infection (ARIs) is an acute respiratory infection that attacks the throat, nose and lungs caused by viruses, bacteria, rickettes. Antibiotics is one way to overcome the disease ARIs. Ineffective use of antibiotics may lead to resistance. The potential value of antibiotics determines the effectiveness of an antibiotic. This study aims to determine the potential value and comparative test of tetracycline antibiotics as a quality evaluation.

Methods: Test the antibiotic potency with diffusion method to use 3 doses with using antibiotic samples. Comparative test of antibiotic testing by comparing samples with standard against *Staphylococcus*

aureus ATCC 29213 test bacteria. **Results:** The potential value obtained was 101.7%, and comparative test was 1 : 1,08. **Conclusion:** The results of testing the potential of this antibiotic in accordance with the requirements contained in Indonesia Pharmacopeia. The comparative test results show that antibiotic samples used had good bacterial killing ability.

KEYWORDS: Tetracycline, antibiotic resistance, potential test, comparative test, agar diffusion.

INTRODUCTION

Basic health research report, the prevalence of ARI in Indonesia is 25.5% (range: 17.5% - 41.4%) with 16 provinces including prevalence above the national figure.^[1] Based on the results of the West Java province's basic health research, the prevalence of ISPA in the city of Tasikmalaya provides a real number for national prevalence.^[2]

ARI can be caused by bacteria, viruses and rickets such as Streptococcus genus, Staphylococcus, Pneumococcus, Hemophilus, Bordetella, and Corynebacterium.^[3] Virus causes include 9 groups Mexovirus, Adenovirus, Coronavirus, Pikornavirus, Mikoplasma, Herpesvirus, and others.^[4]

Testing the potential of antibiotics aims to determine the biological activity of an antibiotic in inhibiting microorganisms, which cannot be determined chemically or physically.^[5] Testing the comparative value of antibiotic activity aims to determine the comparison of antibiotic activity with standard samples against clinically sensitive bacterial isolates.^[5]

In this study will be tested potential test of tetracycline which is also an important data to determine the quality of antibiotics used in the health center of Tasikmalaya City. The requirements of antibiotic levels should be in accordance with Indonesia Pharmacopeia.

MATERIALS AND METHODS

Test Materials

Materials tested were tetracycline standard from PT. Sanbe Farma, Indonesia. McFarland standard No. 0.5, and physiological saline 0.9% (Merck).

Bacteria Test

Test bacteria used to test the potential of tetracycline antibiotics is Staphylococcus aureus ATCC 29213.^[5]

Bacterial Growth Media

Bacteria growth medium used was Mueller Hinton Agar (Merck) with a concentration of 43 g/L and Mueller Hinton Broth (Oxid, Basingstoke, UK) at a concentration of 21 g/L, Mueller Hinton Agar (Merck, USA) with a concentration of 43 g/L.

Method

The antibiotic potency test was performed by agar diffusion method. Determination of antibiotic potency using 3 doses done calculation by formula:

$$I = \log \frac{Dt}{Dm} = \log \frac{Dm}{Dr}$$

$$E = \frac{1}{4} \times [(St - Sr)] + [(Bt - Br)]$$

$$b = \frac{E}{\log 2}$$

$$F = \frac{1}{3} \times [(St + Sm + Sr)] - [(9Bt + Bm + Br)]$$

$$M = \frac{F}{b}$$

$$\text{Potency} = \text{antilog } M \times 100\%.[6]$$

Testing the comparability of tetracycline antibiotics using sample and standard antibiotics with 4 variations of concentration, namely 2 µg / mL, 1 µg / mL, 0.5 µg / mL, and 0.25 µg / mL.^[7]

RESULT AND DISCUSSION

Testing Potential Antibiotics

The potential test of tetracycline antibiotics according to IV Pharmacopoeia Indonesia (1995) was used *Staphylococcus aureus* ATCC 29737 bacteria. The experimental pattern of the agar diffusion method used is the 3 + 3 pattern where one comparison standard is used and one sample with 3 dose variations. The doses used were high doses (0.5 µg / mL), medium doses (0.25 µg / mL), and low doses (0.125 µg / mL).^[6]

The inhibition diameter obtained from the test for the potential of tetracycline antibiotics can be seen in Table 1.

Table 1: Inhibition Diameter of Potential Test Results.

Inhibition Diameter (mm)					
S _{td} H	S _{td} M	S _{td} L	S _{sm} H	S _{sm} M	S _{sm} L
12,2	11,2	10,34	12,3	11,07	10,44

Table Descriptions

S_{td}H : High Dose Standard

S_{sm}H : High Dose Samples

S_{td}M : Middle Dose Standard

S_{sm}M : Middle Dose Medium

S_{td}L : Low Dose Standard

S_{sm}L : Low Dose Samples

The results of antibiotic potency testing of tetracycline with a three-dose pattern obtained for 101.7%. These results indicate that tetracycline antibiotics used in Tasikmalaya city health centers still meet the requirements listed in the Indonesia Pharmacopoeia.^[6]

Antibiotic Comparison Test

The comparative test of antibiotic activity aims to be resistant to antibiotics using resistant clinical isolate bacteria. The result of this treatment is for patients with respiratory infection

(Hermita, 2004). Testing the comparability of tetracycline antibiotics using sample and standard antibiotics with 4 variations of concentration, namely 2 µg / mL, 1 µg / mL, 0.5 µg / mL, and 0.25 µg / mL. The inhibition diameter obtained from the comparative value of tetracycline antibiotic activity can be seen in Table 2.

Table 2: Inhibited Diameter of Antibiotic Comparison Test.

Testing	Inhibition Diameter (mm)			
	2 µg/mL	1 µg/mL	0.5 µg/mL	0.25 µg/mL
Standard	16.8	15.4	12.71	10.87
Sample	16.84	14.71	12.18	10.2

Determination of the value of the appeal of antibiotics is done by making a curve between the log concentration and the inhibitory diameter then a linear regression equation is obtained. After the calculation, the results of the equation for the standard are $y = 6.6837x + 14.861$ with $R^2 = 0.9972$, while the results of the equation for the sample are $y = 7.4577x + 14.605$ with the value $R^2 = 0.998$. The curve between log concentration and inhibition diameter for the standard and sample can be seen in Figure 1, and Figure 2.

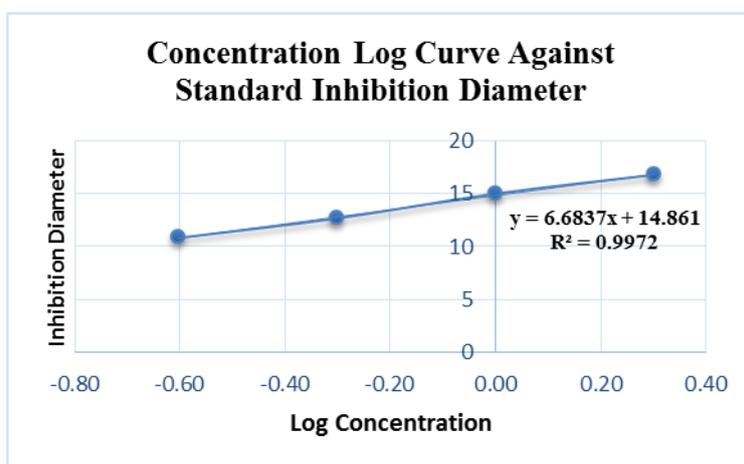


Figure 1: Log concentration curve against standard inhibitory diameter.

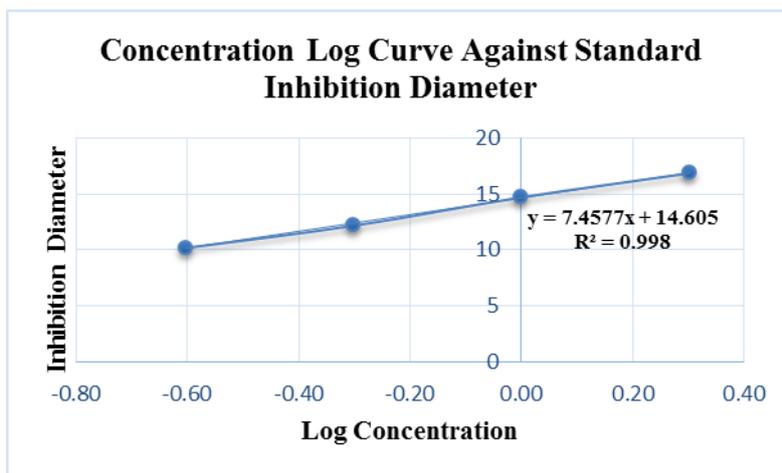


Figure 2: Log concentration curve against sample inhibitory diameter.

The concentration log of the standard antibiotic concentration is substituted as the value of x in the standard equation, so that the value of the inhibitory zone diameter or y value is obtained. Then, the value of y is inserted into the sample equation, so that the value of x is obtained. The antilog value of x is the value of the antibiotic concentration of the sample which is equivalent to the reference standard concentration with the same inhibitory zone diameter. Subsequently, a graph of antibiotic appellate sample values with standard antibiotics was prepared. The graph of the sample and standard antibiotic appellate values can be seen in Figure 3.^[8]

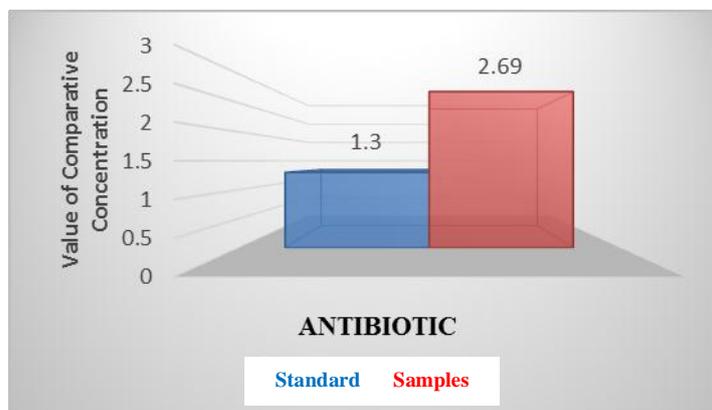


Figure 3: Comparative test values of antibiotic samples and standards.

The comparative value of tetracycline antibiotics obtained is 1: 1.08, this value indicates that the concentration needed by the antibiotic sample is not much different from the standard antibiotic comparison.^[9]

CONCLUSION

The antibiotic potency of tetracycline used in Tasikmalaya City Health Center is 101.7%. Results were still within the range required by the Indonesia Pharmacopeia. The results of comparative antibiotic activity testing showed that the antibiotics used had slightly better activity than the standard with the value 1: 1.08.

ACKNOWLEDGMENTS

The authors are deeply grateful to the subjects participating in this study. The author would like to thank Tasikmalaya City Health Office. The author also thanked Ika Khumairoh for its cooperation in this study.

REFERENCES

1. Ministry of Health of the Republic of Indonesia. 2000. Infant information on ARI. Jakarta: Public Health Counseling Center.
2. West Java Provincial Health Office. Profile of West Java Provincial Health Office. 2003. Bandung : West Java Provincial Health Office. 2003.
3. World Health Organization (WHO). 2007. Prevention and Control of Acute Respiratory Infections (ARI) Tending to Be Epidemic and Pandemic in Healthcare Facilities. America: WHO.
4. Ministry of Health of the Republic of Indonesia. 2002. Guidelines for the Control of Acute Respiratory Disease. Jakarta: Ministry of Health RI.
5. Clinical and Laboratory Standards Institute. Performance Standards for Antimicrobial Susceptibility Testing; Tweny-First Informational Supplement. CLSI document M 100-S 21 (ISBN 1-56238-742-1). Wayne, PA, USA: Clinical and Laboratory Standards Institute; 2011.
6. Ministry of Health of the Republic of Indonesia. 1995. Farmakope Indonesia, Fourth Edition. Jakarta: Ministry of Health of the Republic of Indonesia.
7. Harmita. 2004. Implementation Guidance Validation Method and Method Calculation. Pharmaceutical Science Magazine, December 2004; I(3): 117 - 135.
8. Victor, L. 1980. *Antibiotics in Laboratory Test*. USA: The Williams and Wilkins Company.
9. The United State Pharmacopeial Convention. 2014. *The United States Pharmacopoeia (USP)*. 37th Edition. United States: US Pharmacopeial Convention Inc. 79-82.