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# CERTIFICATE

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as

**Oral Presenter**

A handwritten signature in black ink is written over the ISAC logo.

**ABDI WIRA SEPTAMA PhD**  
**STEERING COMMITTEE ISAC 2020**

**Antimicrobial activity of powder of *Curcuma zedoaria* Roscoe which was stored up to 3 months after irradiation with gamma rays against *Salmonella typhi* and *Escherichia coli*.**

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# Background

- Traditional medicine has been developed in an effort to maintain health and treat disease disorders. One of them is temu putih (*Curcuma zedoaria*) rhizome extract which has antibacterial activity. The use of extract has problem during storage, namely that it is easily damaged by enzymatic processes..



*Fig 1. Curcuma zedoaria Roscoe and Rhizomes*



## PURPOSE

- The purpose of this study was to compare the antibacterial activity of *Curcuma zedoaria* Roscoe (CZR) rhizomes extract which were irradiated with gamma rays and without irradiated against *Salmonella typhi* and *Escherichia coli*. CZR rhizome extract was irradiated with gamma rays at a dose of 0 kGy, 5 kGy, and 10 kGy, then stored until 3 months

*Fig 2. Salmonella typhi and Escherichia coli*



*Salmonella typhi*



*Escherichia coli*

# Co-60 Gamma Ray Irradiation

Irradiation technology (radiation process) is a part of nuclear technology that has developed quite rapidly over the last four decades. Some of these radiation processes have been widely used in various industrial fields and for the preservation of agricultural products, including spices with labels on products with the symbol **RADURA**.



**RADURA**

# METHODS

CZR extract was dissolved in 70% ethanol and dried with a vacuum evaporator, as a comparison were used antibiotics, namely: chloramphenicol and amoxicillin. To find out, whether there were change in the chemical compounds content of the CZR rhizome extraxt due to the effects of irradiation (0 kGy, 5 kGy and 10 kGy), it had been observed the content of the chemical compounds alkaloids, flavonoids, tannins, and saponins. Antimicrobial activity test was carried out by observation of the Inhibitory Zone Diameter by the diffusion method and observation of the MIC by the dilution method.



# RESULT

# Table 1 The Result of Yields

<b>No.</b>	<b>Storage Time</b>	<b>Doses</b>	<b>Initial Powder Weight</b>	<b>Condensed Extracts weights</b>	<b>Yields</b>
1.	0 Bulan	0 kGy	200 g	40,2 g	20,10 %
		5 kGy	200 g	43,9 g	21,95 %
		10 kGy	200 g	47,3 g	23,68 %
2.	3 Bulan	0 kGy	200 g	40,7 g	20,35 %
		5 kGy	200 g	44,5 g	22,25 %
		10 kGy	200 g	47,8 g	23,90 %

# Table 2 Extract Standardization Results

No.	Powder Standardization	Doses (kGy)	Concentration (%)	Indonesian Medika Materia Standards
1.	Water Soluble Content	0	2,829	< 8,9 %
		5	3,145	
		10	3,421	
2.	Soluble Content in Ethanol	0	1,754	< 3,5%
		5	1,844	
		10	1,863	
3.	Ash content	0	3,786	< 4,4%
		5	3,871	
		10	3,145	
4.	Ash content is not dissolved in acids	0	0,229	< 0,74 %
		5	0,191	
		10	0,143	



**Table 4. Bacterial growth rate data  
on Extract Without and With Irradiation (after irradiation)**

<b>Irradiation Dose (kGy)</b>	<b>Number of Bacterial Growth Colonies (cells/mL)</b>
0	$2,5 \times 10^4$
5	0.0
10	0.0

**Table 5. Inhibition Zone Diameter (mm) Activity of Rhizome Ethanol Extract (*Curcuma zedoaria*) Irradiation and without Irradiation during Storage against *Salmonella typhi***

<b>Concentration (%)</b>	<b>0 Month</b>			<b>3 Months</b>		
	<b>0 kGy</b>	<b>5 kGy</b>	<b>10 kGy</b>	<b>0 kGy</b>	<b>5 kGy</b>	<b>10 kGy</b>
<b>12,5</b>	<b>10</b>	<b>12</b>	<b>11</b>	<b>10</b>	<b>12</b>	<b>12</b>
<b>25</b>	<b>13</b>	<b>14</b>	<b>13</b>	<b>12</b>	<b>14</b>	<b>14</b>
<b>50</b>	<b>15</b>	<b>17</b>	<b>17</b>	<b>14</b>	<b>18</b>	<b>17</b>
<b>100</b>	<b>19</b>	<b>20</b>	<b>19</b>	<b>17</b>	<b>22</b>	<b>21</b>

**Table 6. Diameter of Inhibition Zone (mm) Chloramphenicol activity as a comparison to *Salmonella typhi***

Concentration ( $\mu\text{g/ml}$ )	0 Month	3 Months
5	7	6
10	9	9
15	11	11
20	12	12
25	14	14
50	21	22
100	25	25

**Table 7. Diameter of Inhibition Zone (mm) Activity of Rhizome Ethanol Extract (*Curcuma zedoaria*) Irradiated and without Irradiated During Storage against *Escherichia coli***

Concentration (%)	0 Moth			3 Months		
	0 kGy	5 kGy	10 kGy	0 kGy	5 kGy	10 kGy
12,5	11	11	13	10	12	13
25	14	15	15	13	16	15
50	17	18	19	16	19	20
100	19	21	22	19	22	22



**Table 8. Diameter of Inhibition Zone (mm) Amoxicillin activity as a comparison against *Escherichia coli***

Concentration ( $\mu\text{g/ml}$ )	0 Month	3 Moths
5	6	7
10	9	9
20	12	12
25	15	15
30	17	17
50	22	22
100	28	28

**Table 9. Number of growth Bacteria (cells/ml) at MIC of Ethanol Extract (*Curcuma zedoaria*) Irradiated & Without Irradiated During Storage for *Salmonella typhi***

Concentration of Extract in ethanol 10%	0 Month			3 Months		
	0 kGy	5 kGy	10 kGy	0 kGy	5 kGy	10 kGy
2	~	~	~	~	~	~
11	~	~	~	~	~	~
12	$2,88 \times 10^4$	$2,04 \times 10^4$	$2,12 \times 10^4$	$2,8 \times 10^4$	$2,42 \times 10^4$	$2,83 \times 10^4$
13	$2,53 \times 10^4$	$1,96 \times 10^4$	$1,96 \times 10^4$	$2,42 \times 10^4$	$2,17 \times 10^4$	$1,98 \times 10^4$
14	$2,01 \times 10^4$	$1,82 \times 10^4$	$1,32 \times 10^4$	$1,85 \times 10^4$	$1,76 \times 10^4$	$1,55 \times 10^4$
16	$1,17 \times 10^4$	$1,14 \times 10^4$	$9,1 \times 10^3$	$1,26 \times 10^4$	$1,13 \times 10^4$	$8,8 \times 10^3$
18	$8,1 \times 10^3$	$8,7 \times 10^3$	$7,6 \times 10^3$	$7,8 \times 10^3$	$7,3 \times 10^3$	$6,8 \times 10^3$
20	$4,5 \times 10^3$	$3,2 \times 10^3$	$3,8 \times 10^3$	$6,2 \times 10^3$	$6,7 \times 10^3$	$3,4 \times 10^3$
22	$2,4 \times 10^3$	$2,5 \times 10^3$	$2,1 \times 10^3$	$3,2 \times 10^3$	$1,8 \times 10^3$	$1,6 \times 10^3$
24	$1,4 \times 10^3$	$8,0 \times 10^2$	$7,0 \times 10^2$	$2,4 \times 10^2$	$1,1 \times 10^2$	$9,0 \times 10^2$
26	$8,0 \times 10^2$	$8,0 \times 10^2$	$4,0 \times 10^2$	$1,1 \times 10^2$	$6,0 \times 10^2$	$4,0 \times 10^2$
28	$6,0 \times 10^2$	$4,0 \times 10^2$	$1,0 \times 10^2$	$9,0 \times 10^2$	$2,0 \times 10^2$	--
30	--	--	--	--	--	--

**Table 10. Number of Colonies of Ethanol Extract at MIC of Rhizome Extract (Curcuma zedoaria) Irradiated and Without Irradiated during Storage against Escherichia coli**

Concentration in 10% ethanol	0 Month			3 Moths		
	0 kGy	5 kGy	10 kGy	0 kGy	5 kGy	10 kGy
2	~	~	~	~	~	~
11	~	~	~	~	~	~
12	$2,47 \times 10^4$	$2,21 \times 10^4$	$2,11 \times 10^4$	$2,53 \times 10^4$	$2,37 \times 10^4$	$2,03 \times 10^4$
14	$1,74 \times 10^4$	$1,52 \times 10^4$	$1,53 \times 10^4$	$2,03 \times 10^4$	$1,89 \times 10^4$	$1,91 \times 10^4$
16	$1,02 \times 10^4$	$1,18 \times 10^4$	$9,3 \times 10^3$	$1,52 \times 10^4$	$1,17 \times 10^4$	$1,27 \times 10^4$
18	$7,3 \times 10^3$	$6,3 \times 10^3$	$5,8 \times 10^3$	$1,05 \times 10^4$	$8,9 \times 10^3$	$7,6 \times 10^3$
20	$4,2 \times 10^3$	$3,9 \times 10^3$	$2,1 \times 10^3$	$7,9 \times 10^3$	$4,9 \times 10^3$	$3,8 \times 10^3$
22	$2,9 \times 10^3$	$1,4 \times 10^3$	$1,5 \times 10^3$	$5,6 \times 10^3$	$2,8 \times 10^3$	$2,2 \times 10^3$
24	$9,0 \times 10^2$	$9,0 \times 10^2$	$1,0 \times 10^3$	$1,5 \times 10^3$	$7,0 \times 10^2$	$9,0 \times 10^2$
26	$4,0 \times 10^2$	$4,0 \times 10^2$	$3,0 \times 10^2$	$8,0 \times 10^2$	$4,0 \times 10^2$	$2,0 \times 10^2$
28	$1,0 \times 10^2$	$2,0 \times 10^2$	--	$5,0 \times 10^2$	$1,0 \times 10^2$	--
30	--	--	--	--	--	--

# Conclusion

1. The ethanol extract of *Curcuma zedoaria* rhizome irradiated (5 kGy and 10 kGy) and without irradiated (0 kGy) had antibacterial activities against *Salmonella typhi* and *Escherichia coli*. The higher the extract concentration the higher the antibacterial activities.
2. The treatment for 0 kGy, 5 kGy and 10 kGy of extracts storage period for 0 month and 3 months did not affect the antibacterial activities of the extracts.
3. The results of the identification of chemical constituents extract irradiated and the non-irradiated extract was not different
4. The diameter of inhibition zone of the extract at a concentration of 25% against *Salmonella typhi* without irradiated and irradiated at doses of 5 kGy and 10 kGy were average of 14 mm. The same results were obtained for chloramphenicol at a concentration of 25 µg / ml. While the diameter inhibition zone of the extract at a concentration of 25% against *Escherichia coli* without irradiated and irradiated at doses of 5 kGy and 10 kGy were average of 15 mm. The same results were obtained for amoxicillin at a concentration of 25 µg / ml. While, at the Minimum Bactericidal Concentration (MBC) showed no colony growth in both bacteria at all, at irradiated doses and without irradiated either in the 0 month or 3 month storage period, namely at an extract concentration of 30%.
5. For a storage period of 3 months, it was found that there was bacterial growth in the extract that was not irradiated