



## Antibacterial Activity of Some Raw Clays from Congo Brazzaville

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### ABSTRACT

*This work evaluates the antibacterial activity of some raw clays from Congolese localities : Loukolela, Loutete, Missafou, Dolisie and Dongou. Bacterial sensitivity tests were performed on Escherichia coli, staphylococcus aureus and Pseudomonas aeruginosa strains. The raw clays were put in contact with 3 Strains. The results obtained show that all the raw clays simulate the growth of E.coli and P. aeruginosa whereas, that of S. aureus is stimulated essentially by the clays of Loukolela, Loutete and Missafou.*

**Keywords:** Clay; *E.coli*; *P. aeruginosa*; *S. aureus*, Antibacterial.

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### INTRODUCTION

Clay minerals are used therapeutically in the treatment of bacterial infections [1, 2]. It has been proved that the most important way to control diseases caused by bacteria is antibiotic treatment, however, the abuse or overuse of antibiotics causes various side effects such as antibiotic resistance in the body [3, 4]. It is then necessary to develop new types of antibacterial agents as food additives [5]. Some *in vitro* studies have shown that clays protect the intestinal mucosal barrier and absorb toxins, bacterial and viruses [6]. The adsorption capacity of clays is a function of the clay species, 2:1 clays adsorb more than 1:1 clays. In Congo Brazzaville local people in clay areas use clay as a remedy for the treatment of diarrhoea and pregnant women consume it to eliminate nausea caused by pregnancy.

Clays are interesting because they may reveal an antibacterial mechanism, which could provide an inexpensive treatment for bacterial and skin infections, Worldwide [7]. The abundance of clays in Congo Brazzaville would make a low cost for a treatment against bacterial infections. This study is conducted to evaluate the antibacterial activity of raw clays from Congo Brazzaville.

## MATERIALS AND METHODS

The materials used were taken from five localities in Congo-Brazzaville, namely: Loukolela, Loutete, Missafou, Dongou and Dolisie. All these clays were subjected to physicochemical and mineralogical characterization (Table 1) [8-10].

**Table 1: Clay species and origin**

Localities	Predominant mineral	Type
Loukolela	Montmorillonite	2:1
Missafou	Talc	2:1
Loutete	Kaolinite	1:1
Dolisie	Kaolinite and illite	1:1 and 2:1
Dongou	Montmorillonite and kaolinite	2:1 and 1:1

### Clinical bacterial strains tested

The strains used were isolated from patients at the National public health laboratory in Brazzaville and then stored in liquid LB medium plus glycerol in ependof tubes in the refrigerator at -4°C in the laboratory. These strains consisted of two gram-negative bacteria: *Escherichia coli* clinical bacteria and *Pseudomonas aeruginosa* and a gram-positive bacterium *staphylococcus aureus*.

**Culture media used:** The media used are liquid LB medium, Mannitol salt agar, cetrimide agar, Methylene blue eosin and physiological water with 9% NaCl.

**Test for the presence of germs in clays:** A quantity of 7 g of each raw as well as treated unsterilized clay was introduced into five tubes each containing 10 ml of physiological water sterilized at a temperature of 121°C in the autoclave for 15 minutes. 0.1 ml volume of each mixture was plated on agar specific for *pseudomonas aeruginosa*, *Staphylococcus aureus* and *Escherichia Coli*. These cultures were incubated in the oven at a temperature of 37°C for 24 hours.

**Microbial sensitivity testing:** Three bacterial suspensions containing *E.Coli*, *S. aureus* and *P.aeruginosa* strains respectively with a density of 10<sup>6</sup> CFU/ml on the McFarland scale were prepared. 7 g of clay previously sterilized in an autoclave at 121°C were mixed with 10 ml of each of the suspensions. The tubes of mixture thus obtained were incubated after homogenization at 37°C for 24 h. Successive dilutions up to 10<sup>-10</sup> in physiological water were made after incubation for 24 h. A viable colony count was performed and the TSV calculated from the Kra formula [11].

$$TSV = \frac{NA}{NT} \times 100$$
 With NA: number of colonies of the clay-bacteria mixture.

NT: number of colonies in the control.

**X-Ray Fluorescence Analysis:** The chemical analysis was carried out by x-ray fluorescence using a philips X-ray generator. The loss of water was carried out by x-ray fluorescence using a philips x-ray technique is based on the interaction of X-ray radiation with electrons. These interactions lead to the internal reorganization of the electrons, responsible for the emission of a characteristic radiation, which will allow identifying the studied atoms (qualitative aspect). The measurement of the intensity of the characteristic lines will allow to identify the atoms and to determine the centesimal composition of the analysed sample.

## RESULTS AND DISCUSSION

The Table 2 presents the results of the chemical analysis by XRF of the raw clays. It shows that the five clays (Louk, tout, Miss, Doli and Dong) are mainly composed of silica with percentages of 52,96; 60,04; 64,56; 65,93; 55,77 respectively, iron ores with percentages of 7,32 for Louk, 6,00 for Miss, 7,74 for Lout, 3,89 for Dolisie and 3,93 for Dongou and aluminium oxides with 19,05 for Louk, 13,12 for Miss, 30,43 for Lout, 21,76 for Doli, 34,29 for Dong. These results suggest that silica, iron and aluminium ores predominate in the five types of Congolese clays. However, these clays show several differences in their compositions. The percentage of MgO is higher in the Missafou clay than in the others remember that the characterization of miss has revealed an abundance of talc as a clay species. The octahedral layers in talc are constituted of Mg<sup>2+</sup>. Loukolela, Missafou, Dolisie and Dongou on the other hand present a high percentage of iron. We also note that the clays of Loukolela and Dongou have a lower percentage of silica compared to the others.

### Germ Test in Raw Clays

The Table 3 shows the results of the verification test of the presence or not of germs in the different raw clays. The results obtained reveal the presence of *Staphylococcus aureus* and *Escherichia coli* Strains in the clays of Loukolela, Missafou and Loutete while they are totally absent in those of Dolisie and Dongou. The absence of the *Pseudomonas aeruginosa* strain is noted in all the clays. These results show that it is necessary to sterilize these clays before any use.

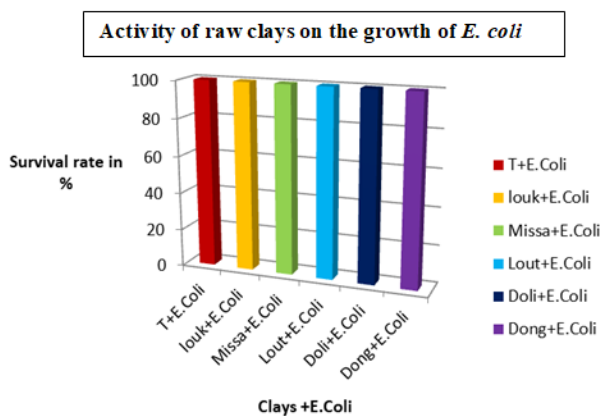
**Table 2: Chemical analysis by X-ray fluorescence of raw clays**

Sample	SiO <sub>2</sub>	Al <sub>2</sub> O <sub>3</sub>	Fe <sub>2</sub> O <sub>3</sub>	MnO	MgO	CaO	K <sub>2</sub> O	Na <sub>2</sub> O	TiO <sub>2</sub>	P <sub>2</sub> O <sub>5</sub>
Loukolela	52,96	19,05	7,32	0,38	2,368	1,87	1,67	0,546	0,72	0,21
Missafou	60,04	13,12	6,00	0,005	6,20	0,20	1,26	0,450	0,54	0,06
Loutete	64,56	30,43	7,74	0,006	0,30	0,11	0,62	0,51	1,68	0,07
Dolisie	65,93	21,76	3,89	13	1,16	0,07	3,16	0,44	1,12	0,10
Dongou	55,77	34,29	3,93	0,34	0,47	0,08	1,066	1,23	2,48	0,07

**Table 3: Germ test in raw clays**

Locality/Strain	Loukolela	Missafou	Loutete	Dolisie	Dongou
<i>Staphylococcus aureus</i>	+	+	+	-	-
<i>Escherichia coli</i>	+	+	+	-	-
<i>Pseudomonas aeruginosa</i>	-	-	-	-	-

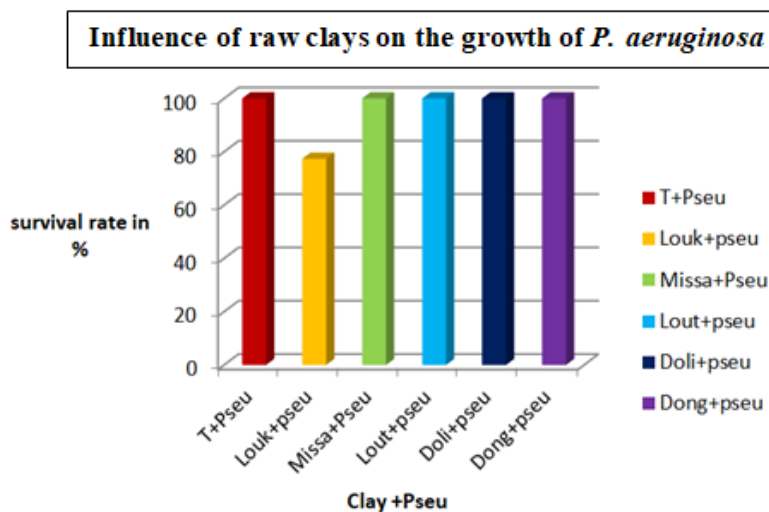
The Figure 1 shows the activity of raw clays on the growth of *Escherichia coli*. It can be seen that raw clays (Louk, Miss, Lout, Doli, and Dong) stimulate the growth of *E. coli* species with a survival rate of 100%. This could be explained by the effect that some microbes in contact with clay minerals draw energy necessary for their growth [12]. These results are close to those kouakou whose clays AK<sub>1</sub>, AK<sub>2</sub>, UB<sub>1</sub> stimulated the growth of *E. coli*.

**Figure 1: Influence of *Escherichia coli* on raw clays**

Note: ■ T+E.Coli; ■ Louk+E.coli; ■ Missa+E.Coli; ■ Lout+E.Coli; ■ Doil+E.Coli; ■ Dong+E.Coli

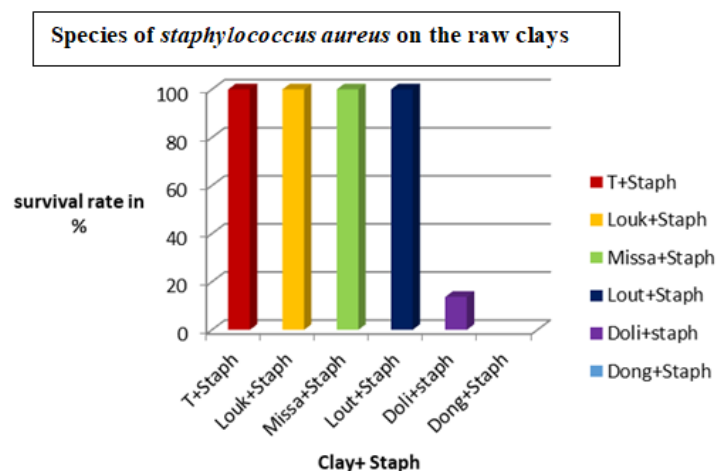
The Figure 2 shows the influence of raw clays on the growth of *P. aeruginosa*. A survival rate of 100% is observed, indicating a possible stimulation of growth. While in the Loukolela clay this growth is lower compared to the clays

in other localities. This activity can be explained by the dominant presence of montmorillonite in Louk [13]. The literature reports that pure and unmodified montmorillonite has no effect against bacteria. A slight decrease in absorbance and number of colonies can be explained by the adsorption properties of the material [14], recall that the characterization of Loukolela clay has shown that the montmorillonite of this clay is of the calcic type, it has been reported in the literature that the  $\text{Ca}^{2+}$  is an essential nutrient for the growth of bacteria [15] since the  $\text{Ca}^{2+}$  in the clay is sequestered in the interfoliar space which favours the slowing down of *Pseudomonas* with the clay and also being a swelling clay, the clay would absorb a large quantity of water from the medium which is unfavourable to the growth of *Pseudomonas aeruginosa*. This result contrasts with that of Kouakou where all clays inhibited the growth of *pseudomonas aeruginosa* species.



**Figure 2: Influence of *Pseudomonas aeruginosa* on raw clays (Note: ■ T+Pseu; ■ Louk+Pseu; ■ Missa+ Pseu; ■ Lout+Pseu; ■ Doil+Pseu; ■ Dong+ Pseu).**

The Figure 3 shows the influence of the species of *staphylococcus aureus* on the raw clays. We note that that the Dongou clay has an inhibitory activity with a survival rate of 0% translating a bactericidal effect and that of Dolisie has a survival rate of (13,66%) showing a bacteriostatic effect on *Staphylococcus aureus*. The clays of Loukolela, Loutete and Missafou stimulate the growth of bacteria. This result is close to that of Kouakou. The strong inhibition observed would certainly come from the presence of illite, montmorillonite, kaolinite in Dongou clay and the combined action of these minerals could induce a decrease in the amount of water in the environment, unfavorable to the development of the species *Staphylococcus aureus* by the type of preponderant minerals and the different combinations.



**Figure 3: Influence of raw and treated clays on the growth of *Staphylococcus aureus***

**Note:** ■ T+Staph; ■ Louk+ Staph; ■ Missa+ Staph; ■ Lout+ Staph; ■ Doil+ Staph; ■ Dong+ Staph

## CONCLUSION

This work evaluates the antibacterial activity of the raw Congolese clays, it emerges from this study that the raw clays of Loukolela, Loutete, Missafou, Dolisie and Dongou are rich in iron, silicon, aluminium, these raw clays stimulate the growth of *Escherichia coli*, *Pseudomonas aeruginosa aureus*. Raw Dolisie and Dongou clays inhibit the growth by showing a bactericidal effect. These results show that some of these clays present an antibacterial activity however this activity can be improved by eliminating certain elements favorable to the development of bacteria. It has been recognized in the literature that the influence of clay minerals on microbial physiological and biochemical properties could both promote and inhibit growth, depending on the microbial type and varying from one species of microorganisms to another. We assume that the physicochemical properties of clays indirectly kill bacteria by producing an unfavorable environment for bacterial growth. These results open a perspective in the use of clays against bacterial infections.

## CONFLICT OF INTERESTS

The author/s declares that they have no conflict of interest.

## REFERENCES

- [1] Fongzossie EF, Tize Z, Nde PF, et al. *South African J Bot.* **2017**;112:29-39. [Cross Ref][Google Scholar]
- [2] Patil SB, Palled MS, Bhat AR. *Asi J Res Chem.* **2010**;3:685-687. [Google Scholar]

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- [3] Ivce R, Sabalja Đ, Zuskin S. Int Symposium ELMAR (ELMAR) **2015**; 15:193-196. [Cross Ref] [Google Scholar]
- [4] Kunin CM. *Annal Internal Med.* **1993**; 118:557-561. [Cross Ref][Google Scholar][PubMed]
- [5] Dong W, Lu Y, Wang W, et al. *Chem Eng J.* **2020**;382:122984. [Cross Ref] [Google Scholar]
- [6] Carretero MI, Gomes CS, Tateo F. *Develop Clay Sci.* **2006**;1:717-741. [Cross Ref][Google Scholar]
- [7] Haydel SE, Remenih CM, Williams LB. *J Antimicrob Chemother.* **2008**;61:353-361. [Cross Ref][Google Scholar] [PubMed]
- [8] Moutou J, Mbedi R, Elimbi A, et al. *Res J Environment Earth Sci.* **2012**;4:316-324.
- [9] Mandimba GR, Mondibaye A. *Biolog Agricult Horticul.* **1996**;13:197-204. [Cross Ref] [Google Scholar]
- [10] Oba RN, Ifo GM, Madila EE, et al. *J Minerals Material Characteriz Engineer.* **2022**;10:93-105. [Cross Ref] [Google Scholar]
- [11] Photos-Jones E, Keane C, Jones AX, et al. *J Archaeolog Sci.* **2015**;57:257-267. [Cross Ref][Google Scholar]
- [12] Morrison KD, Misra R, Williams LB. *Sci Rep.* **2016**;6:1-3. [Cross Ref] [Google Scholar] [PubMed]
- [13] Bartusek S, Skacel A. Int Multidisciplin *Sci GeoCon: SGEM.* **2018**;18:81-87. [Google Scholar]
- [14] Williams LB, Haydel SE, Giese RF, et al. *Clays Clay Min.* **2008**;56:437-452. [Cross Ref] [Google Scholar] [PubMed]
- [15] Brennan FP, Moynihan E, Griffiths BS, et al. *Sci Total Environment.* **2014**;468:302-305. [Cross Ref][Google Scholar] [PubMed]