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Resistance profile of pathogenic strains isolated from community acquired infection at the Protestant Hospital of Ngaoundere (Cameroon)

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ABSTRACT

Worldwide data show that there is increasing resistance among infection bacteria to conventional drugs. The present study aimed at determining data on susceptibility of bacteria isolated from patient for community acquired infection in Protestant Hospital of Ngaoundere against prescribed antibiotics in case of infections by these bacteria. Bacteriological profile of collected samples (stool, urine, cervico-vaginal and urethral secretion, blood) was carry out by standard method. Antibiotic susceptibility was done by the agar disk diffusion method. A total of 120 samples were collected. Among 46 samples was positive for bacterial infections germs (38% of prevalence). Stomach pain represents the most frequent complaint of patients (33.1%). Enterobacteriaceae were the most isolated (70%) followed by *Neisseria gonorrhoeae* (15%). About 73.0% of Enterobacteriaceae identified were resistant to ticarcillin while 80% of *N. gonorrhoeae* to ampicillin. Some of the isolates were resistant to ticarpen (73%), oxacillin (71%) and tobramycin (57%) which are more frequently prescribed and indicates that increased consumption of a particular antibiotic leads to acquisition of resistance. Enterobacteriaceae were predominant bacterial isolated and it is necessary to continue monitoring the consumption of antibiotics to ensure good management of infections.

Keywords: Protestant hospital of Ngaoundere, Bacteriological profile, Bacterial infection.

INTRODUCTION

Infectious diseases have become one of the major health concerns also around the world (Jeanna, 2015). Bacterial infections are responsible for 90% of infections found in health care service (Lacmata et al., 2012). Their diagnosis is essentially based on the isolation of the germ from sample cultures, which remains the key examination. Antibiotics is one way of treating infectious disease which, if it was given irrationally, could lead into a whole new problem, which is

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the presence of a resistant pathogen (Billy, 2003). At the hospital level, knowledge of the main bacterial species responsible for bacterial infection and their level of resistance to antibiotics provides an objective basis for probabilistic antibiotic therapy for these infections (Tamma et al., 2022; Khan et al., 2023). In Cameroon, broad-spectrum antibiotics are commonly used in the hospitals, and there is limited data on antibiotic resistance. The present study was aimed to determine the bacteriological profile and to ascertain the resistance patterns of the most common bacterial isolates from community-acquired infections.

MATERIALS AND METHODS

Study design

A cross-sectional and experimental study carried out on the one hand at the Protestant Hospital of Ngaoundere (PHN), from November 2021 to January 2022. Were included in this study all patients of both sexes, symptomatic, presenting one or more of the following signs or symptoms: abdominal pain, epigastric pain, diarrhea, pelvic pain, urethral discharge, urinary tract problem, associated or not headaches and fevers. Patients on antibiotics treatment were not included in the study.

Isolation and identification of strains from samples

Isolation and identification of different germs were performed using conventional methods of bacteriology (CA-SFM/EUCAST, 2020). The cultural characteristics of agar was used for preliminarily identification through specific staining of colonies on this medium. The confirmatory biochemical identification of germ was done using individual colonies was streaked twice on nutrient agar and characterized through phenotypical tests: colony shape and color, Gram staining, growth at different temperatures and NaCl concentrations, catalase, oxidase, citrate utilization according to Bergey's Manual of Determinative Bacteriology (2005). The API 20 E (BioMérieux SA, France) gallery were used according to the manufacturer's instructions. The specificity of these gallery was at least 92%. The results were recorded and the identification process was performed with Apident 2.0 (BioMérieux, France). Identity of isolates was confirmed using online API web services

Antibiotic susceptibility assay

Antibiotic susceptibility test was done using agar disk diffusion method as recommended by the European Committee on Antimicrobial Susceptibility Testing (CA-SFM/EUCAST, 2020). The antibiotics discs used were strains dependent. Briefly, the inoculum was prepared from a young colony (18 to 24 h old) for each pathogenic strain retained. It was homogenized in 4 mL of sterile saline water (NaCl, 0.85%; *m/v*) and the optical density was adjusted to 0.5 Mac Farland (~ 10^8 cells/mL; OD: 0.08-0.1; wavelength: 620 nm). The antibiotics used: ampicillin, chloramphenicol, ticarpenem, norfloxacin, imipenem, oxacillin, ciprofloxacin, cefotaxime, tobramycin, cefotaxime, erythromycin, rifampicin, minocycline, josamycin, roxithromycine, clindamycin, ofloxacin, clarithromycin, tetracycline, levofloxacine were obtained from Biorad (Biorad, Marnes-la-Coquette, France).

Data collection and analysis

Data was collected from patients 'health books and the bacteriology register of the laboratory. A data collection form was developed with the following variables: sociodemographic (age and sex) and clinical characterizations of patients (symptoms and biological examinations). All data in this work were analyzed using Microsoft Excel and XLSTAT software.

RESULTS

Socio-demographic characteristic of the patients

The present study was conducted to investigate the antibacterial activity on pathogenic strains isolated from community acquired infection at the Protestant Hospital of Ngaoundere. A total number of 120 samples were collected from 120 patients recruited on

the basis of the established inclusion criteria (Figures 1 and 2). Among the 120 patients, 46 were positive for bacterial infections, hence the prevalence rate was 38.8%. Figure 1 shows that the modal age class is [30-40[with 31.7%, while the age group of [60-75[year old constituted the minority with 5.0%. Moreover, from Figure 2 we observed that of the 120 patients, men were predominant (55.0%) compared to women (45.0%) with the sex ratio Men/Women of 1.22.

Clinical characteristics of patients

Figure 3 present the clinical symptoms of patients and in this figure, stomach pain represents the most frequent complaint from patients (33.1%) while chills and hyperthermia constitute the lowest complaint (1.5%). From the Figure 4, a predominance of the examination of the Stool Culture (38.8%), followed by the Urine Culture (30.6%) and the Cervico-vaginal Sample (CVS) (14.9%) is noted. Abscess culture, blood culture, Urethral discharge culture, semen culture and mycoplasma are in the minority, with respective frequencies of 0.8%, 1.7%, 2.5%, 5.0% and 5.8%.

Bacterial content of the samples

The different samples were subjected to a microbiological analysis, in particular for the isolation of pathogenic germs. From the 46 infected samples, 32 (70%) presented Enterobacteriaceae, 7 (15%) *Neisseria gonorrhoeae*, (3) 7% *Staphylococcus aureus* and (4) 9% *Ureaplasma urealyticum* (Figure 5).

Sensitivity of germs to antibiotics (ATBs)

Sensitivity of Enterobacteriaceae

Figure 6 presents the data according to the ATBs tested on Enterobacteriaceae. It generally emerges that of the ATBs tested, Enterobacteriaceae showed respectively 57%, 71% and 73% of resistant to tobramycin (TOB), oxacillin (OXA) and ticarcillin (TIC). On the other hand, they showed a 100% sensitivity to imipenem (IPM).

Sensitivity of *Neisseria gonorrhoeae*

Figure 7 below presents the data according to the ATBs tested on *N. gonorrhoeae*. It generally emerges that of the ATBs tested, isolates showed respectively 40%, 50% and 80% of resistant to oxacillin (OXA), norfloxacin (NOR) and ampicillin (AMP). On the other hand, the showed a 100% sensitivity to imipenem (IPM).

Susceptibility of Staphylococcus aureus

S. aureus was respectively resistant to amoxicillin (AMP) and to oxacillin, but 100%, 50% and 25% (OXA) sensitive respectively to imipenem (IPM), rifampicin (RIF) and erythromycin (Figure 8).

Sensitivity of *Ureaplasma urealyticum*

It generally emerges that of the ATBs tested, *U. urealyticum isolates are respectively* 50%, 50%, 75% and 100% resistant to levofloxacin (LEV), tetracycline (TET), ofloxacin (OFL) and ciprofloxacin (CIP).

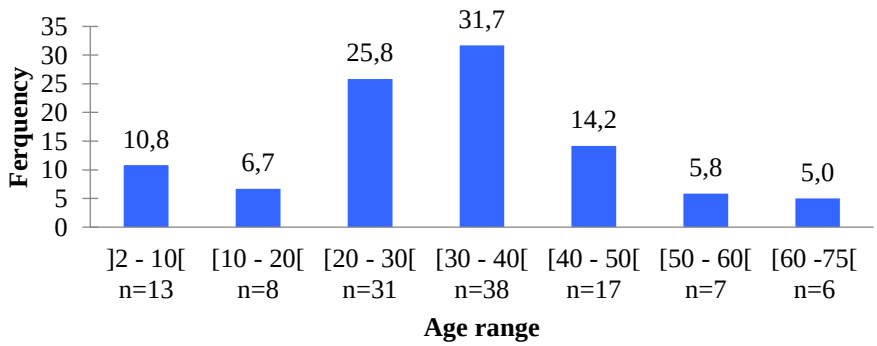


Figure 1: Distribution of patients by age (N=120).

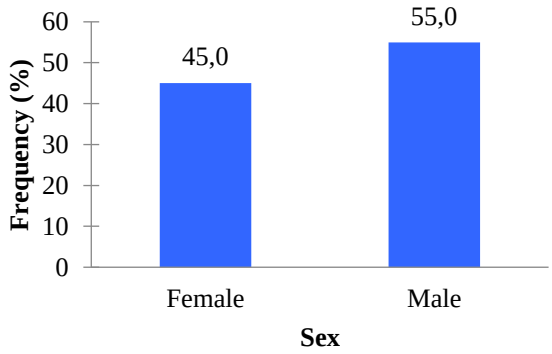


Figure 2: Distribution of patients by gender.

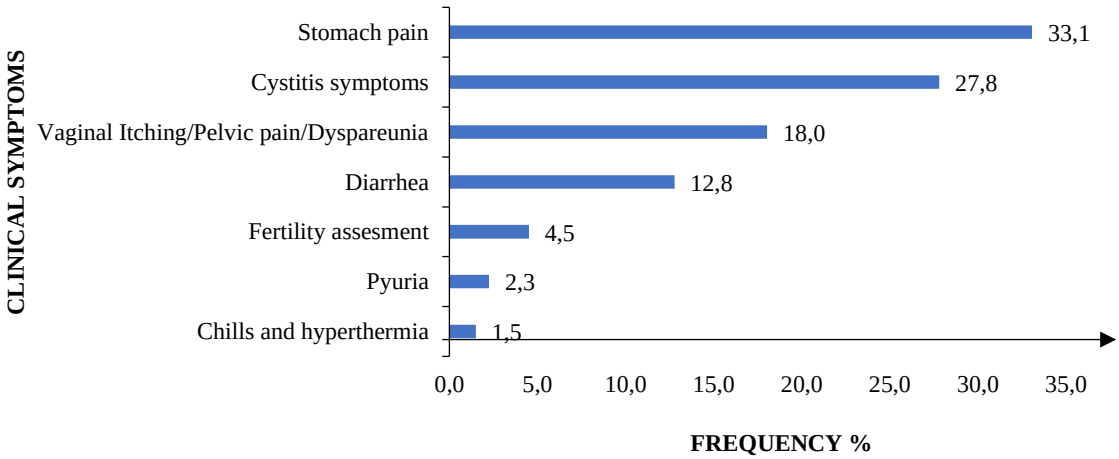


Figure 3: Frequency of clinical symptoms.

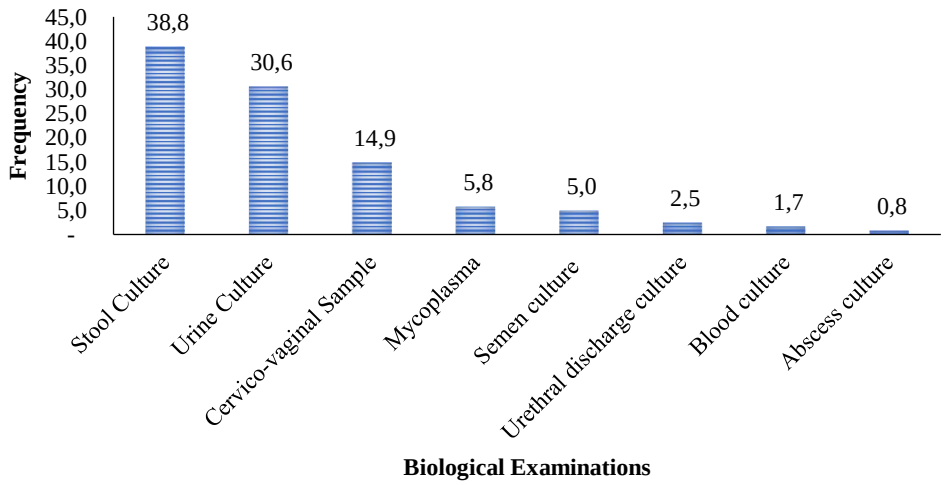


Figure 4: Frequency of biological examinations.

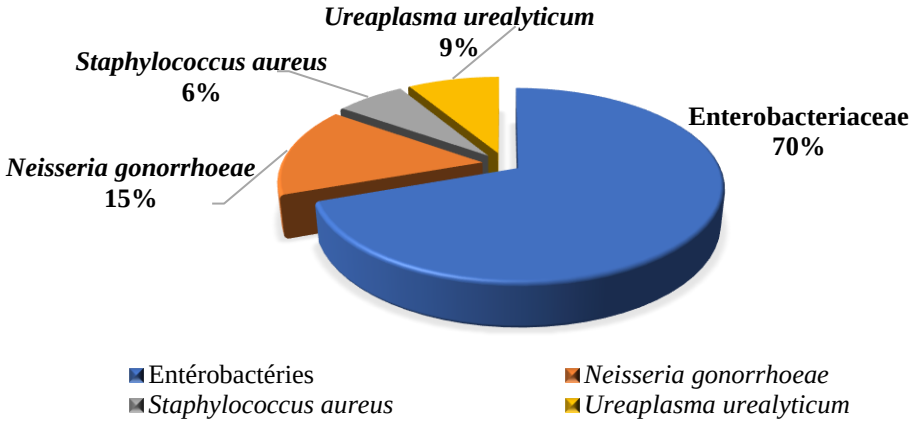
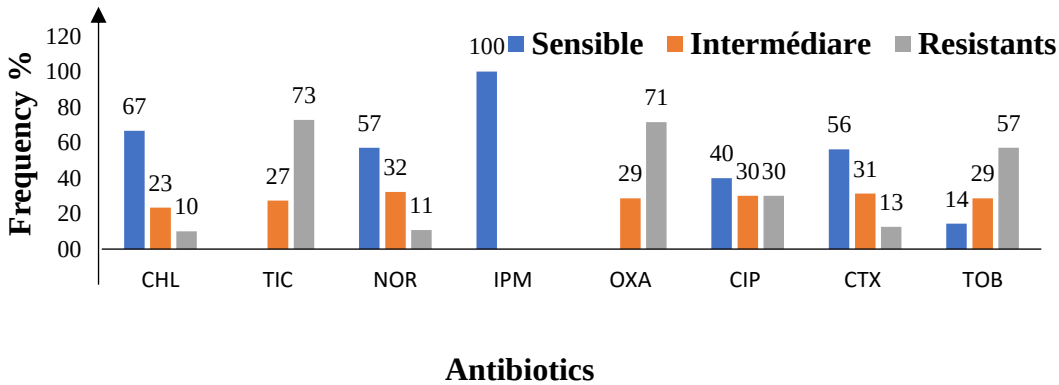
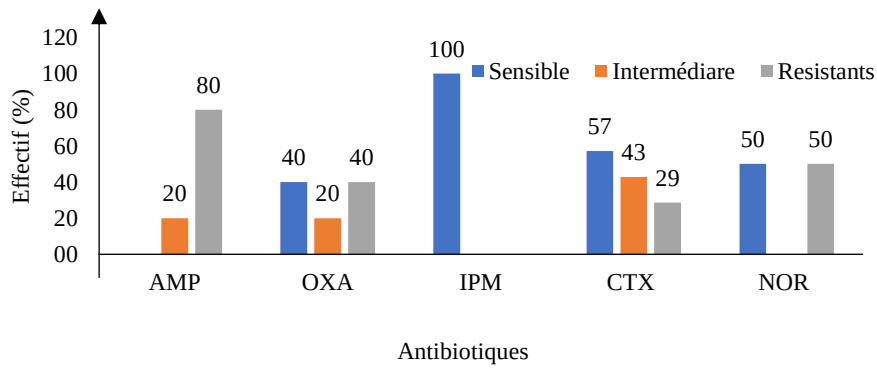


Figure 5: Frequency of pathogenic germs encountered.



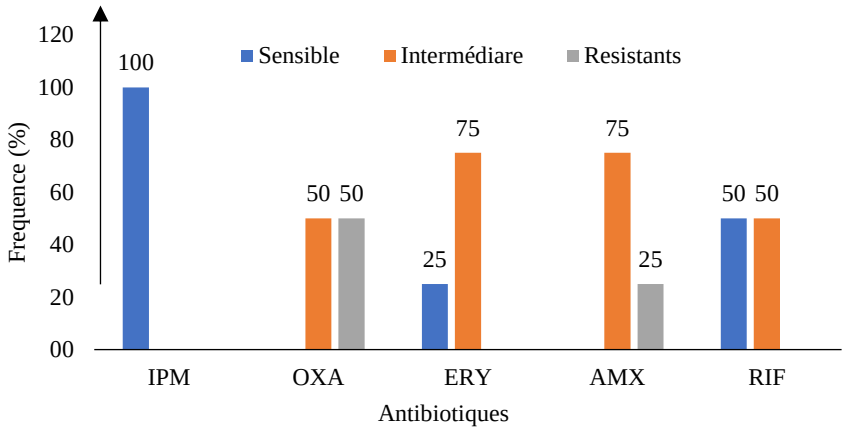
Chloramphenicol (CHL), ticarpen (TIC), norfloxacin (NOR), imipenem (IMP), oxacillin (OXA), ciprofloxacin (CIP), cefotaxime (CTX), tobramycin. (TOB).

Figure 6: Sensitivity of Enterobacteriaceae to antibiotics.



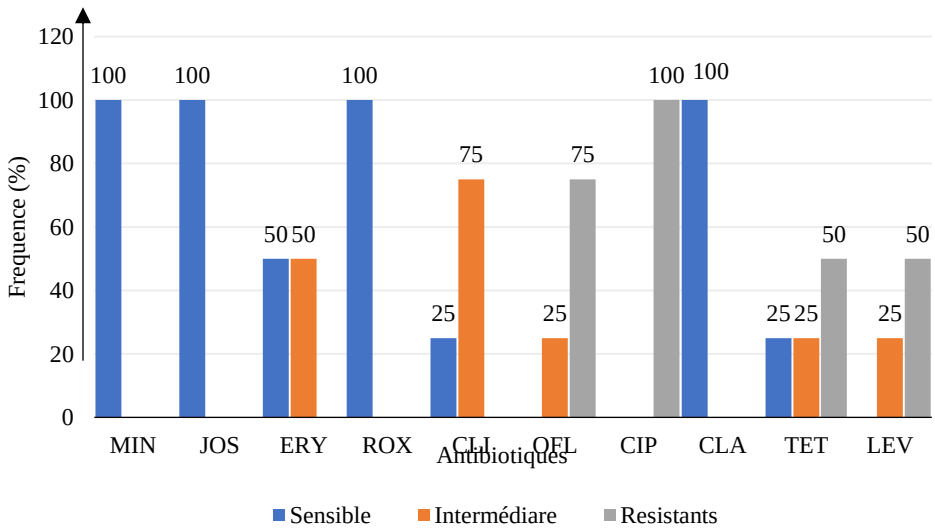
Imipenem (IMP), cefotaxime (CTX), norfloxacin (NOR), ampicillin (AMP), oxacillin (OXA).

Figure 7: Sensitivity of *Neisseria gonorrhoeae* to antibiotics.



Amoxicillin (AMX), imipenem (IMP), erythromycin (ERY), oxacillin (OXA), rifampicin (RIF).

Figure 8: Sensitivity of *Staphylococcus aureus* to antibiotics.



Ciprofloxacin (CIP), erythromycin (ERY), minocyclin (MIN), josamycin (JOS), roxithromycin (ROX), clindamycine (CLI), clarithromycin (CLA), ofloxacin (OFL), tetracyclin (TET), levofloxacin (LEV).

Figure 9 : Sensitivity of *Ureaplasma urealyticum* to antibiotics.

DISCUSSION

Presumptive and adequate antibiotic therapy is essential for the successful treatment of severe infections. This approach requires prior and up-to-date knowledge of the state of bacterial susceptibility to antibiotics. Indeed, the evolution of bacterial susceptibility is characterized by the more or less rapid appearance of new resistance factors. One of the validated strategies for obtaining information on the resistance of bacteria to antibiotics is the continuous surveillance of hospital-acquired and community-acquired infection isolates **(**Ben Jemaa et al., 2004;

WHO, 2021**).** Several publications of CA-SFM / EUCAST (2020) have taken an interest in this subject.

Among the 120 samples collected, the highest frequency (38.8%) was obtained for gastrointestinal infection followed by urinary infection (30.6%), gonococcal infections (15%), *Ureaplasma* – associated infections (9%) and *Staphylococcus aureus* infections (6%). These strains are common in hospitals, known as potential agents of bacterial infections and therefore their ability to resist antibiotic treatments (Cizman, 2003).

These organisms are responsible for various types of infections including, urinary and genital tract infections, dysentery, wound infections, and many others. The high frequency of Enterobacteriaceae could be explained by the fact that, during the present study, most of the participants came to be consulted for abdominal pain. In addition, the multi-resistance of these germs (Gram-) to antibiotics would also justify this frequency (Massongo et al., 2021). This was similar to other studies done in Cameroon for which the most prevalent (77,1%) was *E. coli* (Djim-Adjim-Ngana et al., 2023)

The sensitivity tests of the isolated strains to antibiotics revealed that *N. gonnorhoeae* and Enterobacteriaceae (*Salmonella sp., Shigella spp., Proteus mirabilis, Kluyvera spp.* and *Escherichia coli)* were the most resistant bacteria to antibiotics and showed resistance to the majority of the antibiotics tested. The rate of resistance to third generation cephalosporins and other β-lactam antibiotics can be explained by the high production of penicillinase and the production of extended spectrum β-lactamase (Saïdani et al 2006). However, the rate of this resistance is high compared to those reported in developed countries (WHO, 2021).

Resistance to ticarpen, oxacillin, tobramycin and ampicillin was particularly alarming. This result is consistent with findings from other studies in Yaounde (Nzalie et al., 2016). β-lactams currently remain the molecules most used in the treatment of infections caused by Enterobacteriaceae (Kabrah et al., 2021). However, Enterobacteriaceae naturally harbor and have acquired resistance limiting their activity. These resistances are linked to a lack of accumulation in contact with the target (PLPs or penicillin-binding proteins) following impermeability or efflux of the antibiotic, modifications of the PLPs or the production of inactivating enzymes called β-lactamases (Robin et al, 2012). Imipenem was active against isolated strains and this situation should be preserved. Indeed, we are starting to report strains of Enterobacteriaceae resistant to this molecule which can lead to embarrassing therapeutic impasses. This study revealed a relatively high prevalence of resistance to most antibiotics tested.

Conclusion

In conclusion, the overall objective of this work was to contribute to the identification of the bacteriological profile of samples from patients consulted at the PNH and their sensitivity to antibiotics. The work suggests that imipenem is better for the empirical treatment of bacterial infection in the area. All bacteria isolated showed resistance to more than one drug of choice. In terms of limit, we will mainly retain that it would be good to evaluate the types of resistance (natural or acquired) presented by these germs and the origin of the latter. These limits being underlined, it is appropriate to place this work in a more prospective contest.

COMPETING INTERESTS

The authors declare that they have no competing interests.

AUTHORS' CONTRIBUTIONS

TTB and DHD designed the study. TTB supervised, validated, writing reviewed and edited. DHD, SYN and DEX isolated and identified the strains at the hospital. DTCL and HDRC made the extracts from the plants. NSFS, TTB and DHD analyzed, interpreted the data and wrote the first draft. TTB, NSFS, DHD and DTCL reviewed the manuscript. All authors have read and approved the final version of the manuscript.

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We are grateful to all the staff of the laboratory of the Protestant Hospital of Ngaoundere for their collaboration as well as all the patients who accepted to participate in this study.

Ethical considerations

We obtained a research certificate from the University of Ngaoundere as well as a research authorization from the Protestant Hospital of Ngaoundere to get required data. Identification codes were used for data treatment to ensure confidentiality and privacy of information; results were used solely for scientific purposes.

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