BioRisk 15: 31–43 (2020)
doi: 10.3897/biorisk.15.58455
https://biorisk.pensoft.net

RESEARCH ARTICLE

A peer-reviewed open-access journal
BioRisk

Total and faecal coliforms presence in cenotes of Cancun; Quintana Roo, Mexico

Jessica Borbolla-Vazquez[1], Paul Ugalde-Silva[2], José León-Borges[3], Job Alí Díaz-Hernández[1]

1 *Polytechnic University of Quintana Roo, Cancun, Mexico* **2** *CINVESTAV-IPN, Mexico, Mexico* **3** *University of Quintana Roo, Cancun, Mexico*

Corresponding author: Job Alí Díaz-Hernández (job.diaz@upqroo.edu.mx)

Academic editor: J. Settele | Received 8 September 2020 | Accepted 14 November 2020 | Published 2 December 2020

Citation: Borbolla-Vazquez J, Ugalde-Silva P, León-Borges J, Díaz-Hernández JA (2020) Total and faecal coliforms presence in cenotes of Cancun; Quintana Roo, Mexico. BioRisk 15: 31–43. https://doi.org/10.3897/biorisk.15.58455

Abstract

The large increase in population in Cancun, Mexico has increased domestic, agricultural and industrial activities, resulting in inadequate solid and liquid waste management that can affect underground aquifers. One of the factors which affects water quality is coliform bacteria. The present study focused on determining the presence of total and faecal coliforms in ten urban cenotes in Cancun. Sampling was carried out in the dry and rainy seasons of 2018. The Most Probable Number (MPN) technique was used to determine the concentration of coliform bacteria. The results from the analyses indicate that the ten cenotes are contaminated with total and faecal coliforms. Additionally, the concentration of coliforms increases during the rainy season. We conclude that all the cenotes are contaminated with faecal coliforms and suggest that more studies are necessary to determine the origin of this contamination and the impact on the ecosystem.

Keywords

cenote, faecal coliform, total coliform, underground water

Introduction

Cancun is located north of Quintana Roo State, which is one of the thirty-two federal entities of Mexico. The geographical coordinates of Cancun are 21°09'41"N, 86°49'29"W, with an altitude of 5 m above sea level. This city has a humid subtropical climate with summer rainfall and its average temperature ranges from 28 °C to 30 °C in the warmest months (from April to August). The rainy season in Cancun usually starts in August and ends in October, with an average rainfall of about 200 mm; however, in the past few years, the rainy season has extended until November [Instituto Nacional de Estadística y Geografía (INEGI) 2017].

The City is built on an extensive plain formed by calcite layers derived from the deposition of calcareous minerals (mainly calcium or magnesium carbonate) from shells and exoskeletons of marine organisms. The exposure of these calcite layers to underground aquatic currents formed by rainfall and river formation generates the calcite dissolution. Over time, the constant erosion of these layers generates depressions, holes and caverns, which produce peculiar structures called "*cenotes*", that are present throughout the Yucatan Peninsula (Suárez-Morales and Rivera-Arriaga 1998; Escolero et al. 2005). The Yucatan Peninsula has suffered multiple fractures and fissures that allow water filtration and thus prevent the formation of aquatic bodies on the surface. Therefore, most of the water for human use in the state of Quintana Roo is contained in aquifers which constitute the only source of freshwater for human settlements (Escolero et al. 2005; Nava-Galindo 2015).

Since its foundation in 1971, Cancun has experienced an exponential population growth. In 2010, there were 628306 inhabitants, while in 2015, the population increased to 743626 inhabitants (INEGI 2017). The rapid development and growth that the city has suffered raises the question about its capacity to develop sustainable use of its natural resources [Secretaría de Turismo (SECTUR) 2013; INEGI 2017]. The increase in domestic, agricultural and industrial activities has resulted in inadequate management and control of solid and liquid wastes, thus affecting the quality of aquatic ecosystems, mainly in aquifers. This overproduction of waste from human activities has, therefore, caused ecological, socioeconomic and health problems (Ramos et al. 2008). This situation has decreased the availability and quality of freshwater for human use and has increased the expenditure for energy to obtain it. To address these problems, some alternatives have been proposed, such as obtaining freshwater from alternative reserves or expensive purification processes (Hoogesteijn-Reul et al. 2015; Nava-Galindo 2015).

One of the biological factors that affects water quality is the presence of coliform bacteria. Total coliform bacteria are harmless microorganisms which live in the intestines of man and warm and cold-blooded animals, where they help digestive processes. A small group of this type of bacteria has been classified as faecal coliform bacteria, where the most common member is *Escherichia coli*. This type of bacteria has been separated from the total coliform group because of its ability to grow at elevated temperatures and its relationship with faecal material from warm-blooded animals. This group is mainly composed of gram-negative bacilli, which are not sporulated, oxidase-negative, aerobic or anaerobic facultative, are able to multiply in the presence of bile salts and are able to ferment lactose with acid and gas production in 48 h at 44 °C (Mossel 1982). As faecal coliform bacteria, such as *E. coli*, are generally not pathogenic, they are used as organisms to indicate the presence of pathogenic bacteria. These indicator bacteria are always present in the same gastrointestinal tract of organisms infected with pathogenic bacteria, but the latter are in such small quantities that it is not practical to monitor them directly (Rochelle-Newall et al. 2015).

Usually the presence of faecal coliform bacteria is an indicator of water contamination. The presence of these bacteria in underground water is generally related to outbreaks of virulent diseases like cholera, dysentery, paratyphoid fever, hepatitis, stomach infection and septicaemia. In tropical and under-developed cities, these diseases are transmitted by faecal contamination of underground water [Ogbondeminu et al. 1994; Pacheco et al. 2000; McDonald et al. 2008; Norma Mexicana NMX-AA-042-SCFI-2015 (2015)]. Coliform studies on urban cenotes are scarce; the most studied are tourist cenotes. Therefore, the present study is focused on determining and analysing total and faecal coliforms' concentrations in Cancun's cenotes, which are used as natural swimming pools and freshwater sources.

Material and methods

Sampling

The samples were collected from ten cenotes located in Cancun, one sample from each cenote. The sampling was carried out in two seasons of the year 2018, from May to June (dry season) and from September to October (rainy season) (Fig. 1 and Table 1). The samples were collected in the morning (from 9 to 10 am), in the middle of the cenote at 10 cm under the water surface and collected in 500 ml sterile screw cap glass bottles by filling them to 3/4 of their capacity. The glass bottles were labelled and kept at 4 °C until their transfer to the Biotechnology Laboratory at the Polytechnic University of Quintana Roo (UPQRoo), in a period not exceeding 2 h after sampling.

Sample analysis

The determination of total and faecal coliform was carried out by the technique of Most Probable Number (MPN), according to the Mexican Norm NMX-AA-042-SCFI-2015 and the American Public Health Association (APHA), American Water Works Association and Water Environment Federation (WEF) (1998). In the case of total coliform bacteria, 10, 1.0 and 0.1 ml of the samples were taken and added to lactose-rich MCD liquid medium tubes with an inverted fermentation Durham´s tube inside. Double lactose concentration was used for tubes with 10 ml of sample, while normal lactose concentration was used for tubes with 1.0 and 0.1 ml samples. The tubes were incubated at 37 °C for 48 h. After the incubation period, the tubes that presented turbidity and the presence of gas collected in the Durham´s tubes were considered positive for total coliforms. In the case of faecal coliform bacteria, the positive total coliform samples were grown in tubes containing Brilliant Green Bile Lactose liquid medium (Bioxon) and an inverted fermentation Durham´s tube. The tubes were incubated at 45 °C for 24 h. After the incubation period, the tubes that presented tur-

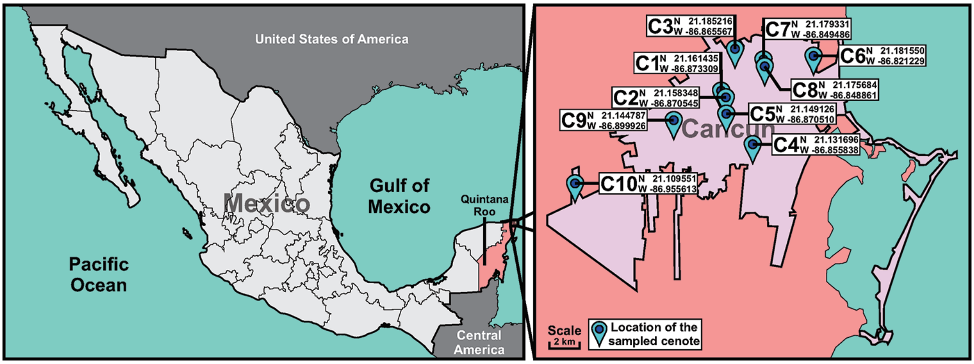

Figure 1. Location of the sampled cenotes in Cancun, Quintana Roo, Mexico. On the left, the location of Quintana Roo State in Mexico; on the right, blue markers (C1-C10) correspond to the global coordinates of each cenote; in lilac is the Cancun urban area.

bidity, the presence of gas collected in the Durham´s tubes and a slight change in the colour of the medium were considered positive for faecal coliform bacteria.

The Most Probable Number (MPN) of total and faecal coliform bacteria contained in 100 ml of the sample was calculated by comparing the number of positive tubes in the confirmatory results with statistical tables listed in the Mexican Norm NMX-AA-042-SCFI-2015 (2015). The data were listed and plotted.

Proximal analysis

The distances between cenotes were obtained using the Haversine formula (Robusto 1957).

$$d = 2r\,sin^{-1}\left(\sqrt{sin^{2}\left(\frac{\grave{e}_2-\grave{e}_1}{2}\right)+\left(\cos\left(\grave{e}_1\right)*\cos\left(\grave{e}_2\right)*sin^{2}\left(\frac{\tilde{a}_2-\tilde{a}_1}{2}\right)\right)}\right) \quad (1)$$

Euclidean distance was used for "nearest" criteria; in this work, the quantity spheric polygon centroid is inferred using the following formula:

$$\mathrm{dist}(x,y)=\sqrt{\sum_{i=1}^{n}\left(x_i-y_i\right)^2} \quad (2)$$

Cluster analysis

For the process algorithm, formulated by MacQueen (1967), first, we have partitioned the data in clusters, denominated as "k records"; second, we randomly assigned "k records" to be the locations of the cluster centres for each k record, to find the nearest cluster centre. Thus, each cluster centre "owns" a subset of the k records, thereby representing a partition of the dataset. Thus, we have "k clusters, C1, C2, …, C*k*" for each

Table 1. Concentrations of total and faecal coliform bacteria.

Sampled cenote	Total coliform		Faecal coliform	
	Dry season	Rainy season	Dry season	Rainy season
	May–Jun	Sep–Oct	May–Jun	Sep–Oct
C1	93	> 2400	93	240
C2	> 2400	253	> 2400	253
C3	240	> 2400	240	157
C4	1100	> 2400	1100	> 2400
C5	> 2400	> 2400	> 2400	> 2400
C6	> 2400	> 2400	> 2400	110
C7	< 3	> 2400	< 3	157
C8	< 3	> 2400	< 3	> 2400
C9	93	253	4	79
C10	21	> 2400	7	> 2400

* Data are expressed by the most probable number of total or faecal coliform bacteria in 100 ml of sample (MPN/100 ml).

of the *k* clusters. Finally, we need to find the cluster centroid and update the location of each cluster centre and determine the new value of the centroid. We repeat this process until there is convergence or termination.

Clustering analysis is performed through the distance obtained in latitude and longitude. To make this analysis, three groups were carried out, based on latitude and longitude distances, considering the closest in each of the groups. The number of the resulting groups of equal latitude and longitude are placed in the final group; on the other hand, the number of the resulting groups of latitude and longitude that are not equal are labelled as the missing groups. Considering the map of Cancun, the result is four cenotes to the northeast, one to the southwest and five at the centre. Then the *spherical polygon centroid* function of Robert J. Hijmans is used, based on the formula (Williams 2011) *Rhumb Line Navigation* contained in the geosphere package (Hijmans et al. 2017) of the statistical analysis programming language R. The result is plotted through the QGIS software.

Results

For processing and analysis of data, cenotes were named arbitrarily with the code C1 referring to the first sampled cenote and so on until C10 referring to the tenth sampled cenote (Fig. 1). In the dry season, three cenotes had the highest concentration (> 2400 MPN/100 ml) of total coliforms; the same cenotes had the highest concentration of faecal coliforms in this season. In the rainy season eight cenotes had the highest concentration (> 2400 MPN/100 ml) of total coliforms, while four cenotes had the highest concentration of faecal coliforms in the same season (Table 1).

To understand the changes in the concentration of coliforms in cenotes between both seasons, the frequencies of each concentration of the Most Probable Number (MPN/100 ml) of total (Fig. 2A) or faecal coliforms (Fig. 2B) were plotted. During the dry season, only 3 cenotes had > 2400 MPN/100 ml and 1 had 1100 MPN/100ml

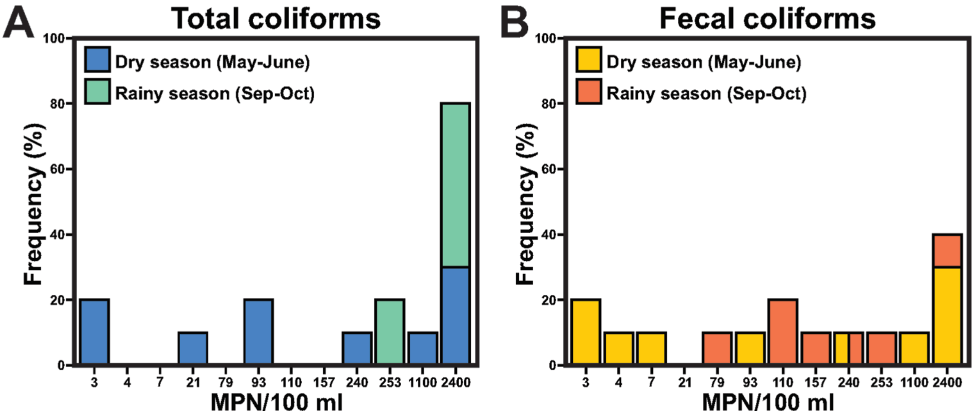

Figure 2. Relative frequencies of concentrations of total and faecal coliforms in cenotes of Cancun **A** frequencies of different concentrations of the Most Probable Number (MPN/100 ml) of total coliforms detected during dry (May-June) and rainy (September-October) seasons **B** frequencies of different concentrations of the most probable number (MPN/100 ml) of faecal coliforms detected during dry (May-June) and rainy (September-October) seasons. Frequency is plotted as a percentage of cenotes with the same concentration of MPN/100 ml. Bars with the same percentage but with both colours represents cenotes without changes between dry and rainy seasons.

of total coliforms; according to the Mexican norm the limits are <200 MPN/100 ml to swim, these cenotes considered to have high concentrations of bacteria. In contrast, the other 6 of cenotes had low concentrations of total coliforms, which ranged from 3-240 MPN/100 ml; however only 5 cenotes have concentrations below the limits of the Mexican norm. Interestingly, in the rainy season, the number of cenotes with high concentrations (> 2400 MPN/100 ml) of total coliform bacteria increased to 8 and only 2 had low concentrations (253 MPN/100 ml) of total coliform bacteria (Fig. 2A). In this season, all cenotes exceed the limits of the Mexican norm.

On the other hand, in the dry season, faecal coliforms were detected in high concentrations (> 2400 and 1100 MPN/100 ml) in 3 and 1 of the cenotes, respectively, while the remaining 6 of the cenotes had low concentrations of faecal coliforms, ranging between 3 and 240 MPN/100 ml. In the rainy season, 4 of the cenotes had high concentration of faecal coliforms (> 2400 MPN/100 ml) and the other 6 cenotes had low concentrations of faecal coliforms (ranging from 79 to 253 MPN/100 ml) (Fig. 2B). Therefore, it is evident that the concentrations of total and faecal coliforms increase in the rainy season.

The Yucatan Peninsula has the largest underwater cave system in the world (Bauer-Gottwein et al. 2011) and that many of these caves are interconnected by an extensive hydrological system (Gómez-Nicolás et al. 2017). To determine the possible hydrological connection of the ten sampled cenotes, an analysis of proximity of cenotes was performed (Fig. 3). Three clusters (CL1, CL2 and CL3) were obtained from this analysis. Each cluster has a centroid, which represents the geographic centre and is surrounded by a minimum (radius of 2 km), a medium (radius of 2.5 km) and a maximum (radius

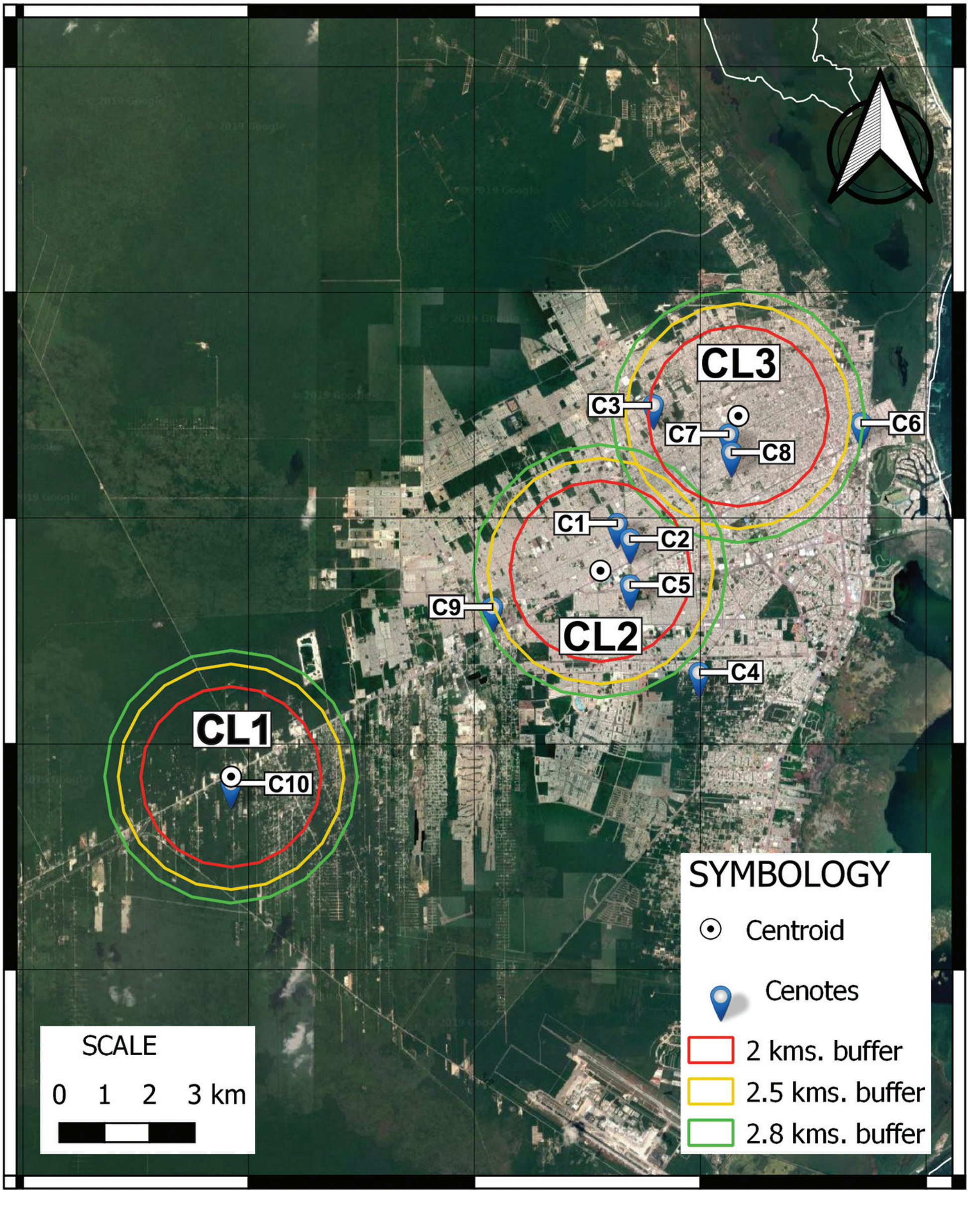

Figure 3. Cluster of cenotes of Cancun; each cluster (CL) has a centroid with a minimum (red circle with a radius of 2 km), a medium (yellow circle with a radius of 2.5 km) and a maximum (green circle with a radius of 2.8 km) circular perimeters. In CL1, only cenote C10 is located; in CL2, C1, C2 and C5 cenotes are located and in CL3, C3, C7 and C8 cenotes are located.

of 2.8 km) circular perimeter. C1, C2 and C5 cenotes were located within the minimum circular perimeter of CL2. A similar pattern was detected in CL3, where C3, C7 and C8 cenotes were detected near the centroid and C6 and C9 cenotes were detected at the maximum circular perimeter of CL3 and CL2, respectively. Finally, in CL1, only C10 was found. It should be noted that this is a predictive analysis by the computer due to its distance. However, to prove if the cenotes are connected, a more accurate and geographical analysis is required.

Discussion

Coliform bacteria are a very diverse group with several members. Some of these members are *Escherichia coli*, *Salmonella sp*., *Klebsiella sp*., *Enterobacter sp*., *Citrobacter sp*. and *Shigella sp*. According to data from the World Health Organization (WHO) (2004), contamination of food or drinking water with coliform bacteria is a risk to human health (Casanovas-Massana and Blanch 2013; Bojarczuk et al. 2018).

It has already been demonstrated that the presence of faecal coliforms is closely related to human activities. A study conducted by McDonald et al. 2008 reported that, in 480 samples taken in the New Cairngorms National Park in Scotland, 85% were positive for total coliform bacteria. It should be noted that the sampling was taken in the four main rivers of the Park where there is a camping area. The authors concluded that, when the number of visitors increases, the probability of identifying faecal coliform bacteria, such as *E. coli*, also increases.

An and Breindenbach (2005) demonstrated the presence of total coliforms in all samples collected from spring waters in Seoul, South Korea. They demonstrated that *E. coli* was detected in 68% of the samples. They also found that all samples located near the public spring baths contained high concentrations of *E. coli*. These data demonstrate the importance of continuous monitoring of water bodies for maintaining a biological balance in order to avoid harm to human health and other organisms.

In Mexico, Pacheco et al. (2000) determined the bacterial contamination of groundwater in a rural region of Yucatan, Mexico. They identified high concentrations of bacteria, such as *E. coli*, *Serratia sp*. and *Enterobacter sp*., which remarkably exceeded the maximum permissible limits established by the Mexican Official Norm. Therefore, since these bacteria are causes of severe gastroenteritis, the authors suggested that continuous monitoring of groundwater for human and cattle consumption be undertaken.

In the present study, samples were taken from urban environments (except cenote 10, which is located in a semi-urban context with many random human settlements). All other cenotes are located within Cancun. The results show that all sampled cenotes are contaminated with total and faecal coliforms to a greater or lesser extent and that the concentration of both coliform bacteria increases in the rainy season in several cenotes (Table 1). Thus, these observations indicate that rain is an important factor that increases the total and faecal coliform concentrations in most cenotes. This research agrees with other reports (Kane 2016; Rosiles-González et al. 2017), the authors show that levels of total coliforms or E. coli increase in the rainy season in 3 cenotes in Cancun; of them, we analysed one cenote that was named C2 in our study.

We suggest that the change in the concentrations of total and faecal coliforms during the rainy season could be related to several factors. The first factor can be anthropogenic activity in the heavily populated areas, where we find a lot of garbage and the increased flow of surface water in the rainy season flushes the waste into underground water system, thus causing pollution in the cenotes. It is also known that some illegal settlements use latrines and septic tanks whose contents will be flushed to the drainage system during times of heavy rainfall and thereby infiltrate to the groundwater or cenotes due to the porosity of the ground in Cancun. Previous research groups also demonstrated that there is a directly proportional relationship between the presence of anthropogenic activities and faecal coliform bacteria (Bauer-Gottwein et al. 2011; Derrien et al. 2015; Moore et al. 2020). Thus, pollution levels in the underground waters of Cancun should be constantly monitored to avoid diseases related to faecal coliform bacteria. It is important to emphasise that, in most of the rural regions of the Yucatan Peninsula, the main sources of freshwater are groundwater found in cenotes [Comisión Nacional del Agua (CONAGUA) (2015)]. We found that all the analysed cenotes were contaminated. We suggest that there is a high probability that the aquifer under this city is contaminated. This hypothesis is supported by our computational analysis of proximity (Figs 2, 3), and by the study of Leal-Bautista et al. (2019) who found total and faecal coliforms at the groundwater source (wells) located west of Cancun.

Data from the computational analysis of proximity suggest that the cenotes inside CL2 (C1, C2 and C5) and CL3 (C3, C7 and C8) have hydrological connections that could influence changes in total and faecal coliform concentrations (Fig. 3). On the other hand, CL1 only has one cenote; C10 and C4, C6 and C9 cenotes are not close enough to each other for possible hydrological connections. This analysis showed that at least seven of the ten sampled cenotes could be connected. Moreover, C5 and C6 cenotes are the most contaminated of the sampled cenotes in the dry and rainy seasons. It should also be noted that there are approximately 150 cenotes registered within the locality of Cancun according to the data provided by the region's Department of Ecology. Therefore, there is a very high probability that there are more underground connections between the cenotes.

As there is a high probability that each of these cenotes is connected to a large aquifer, it is very likely that soon all of these sources of underground water will also be contaminated. Interestingly, cenote C9 is the least contaminated of all the cenotes sampled in both seasons. This could be the result of a wooded area existing above the cenote which could be preventing the filtration of contaminated water into the cenote. However, in the rainy season, the concentration of total and faecal coliforms decreases in cenote C2, perhaps due either to a flow of underground water that goes from cenote C2 to another location, or the source of coliform contamination was eliminated as a result of the municipal programme for cleaning the garbage from the cenotes. This programme, however, does not exist in all areas in Cancun. Therefore, tracers are necessary to identify the source and destination of these contaminants.

On the other hand, *E. coli* is the most representative bacteria of faecal coliforms that affect human health (An and Breindenbach 2005; McDonald et al. 2008). Nevertheless, this bacterium is not the only faecal coliform that can affect human health. For this reason, this study focused on the general presence of faecal coliforms that usually inhabit the digestive tract. According to this, in the Cancun-Tulum tourist corridor, faecal coliforms have been detected in five cenotes used for tourist activities. Concentrations of 0-460 MPN/100 ml of faecal coliforms were detected in four of them and one of them exceeded 1000 MPN/100 ml (Alcocer et al. 1998). A study determined faecal contamination in 48 cenotes used for tourist activities in the State of Yucatan. They found that all cenotes tested positive for the presence of total and faecal coliforms, indicating that they were contaminated with faecal matter (Hoogesteijn-Reul et al. 2015). Furthermore, the permissible limits of total and faecal coliforms established by the Mexican Norm NMX-AA-042-SCFI-2015 (2015) were exceed. Although coliforms are responsible for many diseases and doctors have reported illness from swimming in cenotes, nobody has yet directly sampled the cenotes in connection with determining coliform bacteria´s presence and relate the presence of these bacteria or other organisms with these diseases.

The authors strongly recommend that the Government should consider a sanitation policy for the use of cenotes as tourist/recreational centres. Therefore, the use of cenotes as swimming pools presents an adverse environmental impact since they do not have sanitation systems developed for them. The coliform contamination of cenotes also affects the balance of the microenvironment by altering the food web. We have found that, in some cenotes with high levels of faecal coliforms, alteration in the population of some species of microalgae and protozoa species exists (unpublished data). In addition to the environmental problems, there is an economic and health risk because the underground water of the cenotes is the only source of freshwater in the Cancun Region. This fact implies that freshwater for human use has to be extracted from distant locations, thus increasing the cost of the process.

Conclusion

The concentrations of total and fecal coliforms from 10 cenotes located within Cancun were monitored in two seasons of the year, dry and rainy. Levels from 93 to 2400 MPN / 100 mL of total coliforms were detected in the dry season (3 cenotes had the maximum detectable > 2400 MPN / 100 mL), in the rainy season 9 cenotes showed an increase in concentration, from which 8 cenotes had the maximum detectable. While in 7 cenotes faecal coliforms concentration increased in a range from 79 to 2400 MPN / 100 mL, in the rainy season. In such a way, the rain is the main factor that increases coliforms concentrations in the cenotes.

This pollution in urban cenotes implies a serious problem for the management of residual wastewater in Cancun, and could adversely affect the nearby ecosystem, like lagoons, estuaries and coral reefs, causing a serious deterioration of this ecosystem and in public health. Additional studies and continuous monitoring are necessary. Therefore, it is urgent to implement severe restrictions for household waste, as well as to increase ecological education for the care of cenotes. As many of these sites are used as garbage dumps, the current situation is already serious and causing damage that could be irreparable and costly in the long term.

Acknowledgements

Thanks to Polytechnic University of Quintana Roo, special thanks to Dr. Iris Nava for academic support, Ing. Tania Montoya for technical support and Mario Alberto Luna-Pavo for technical support in software QGis. This research did not receive any specific grant from funding agencies in the public, commercial or not-for-profit sectors.

References

Alcocer J, Lugo A, Marín L, Escobar E (1998) Hydrochemistry of waters from five cenotes and evaluation of their suitability for drinking-water supplies, northeastern Yucatan, Mexico. Hydrogeology Journal 6: 293–301. https://doi.org/10.1007/s100400050152

An YJ, Breindenbach GP (2005) Monitoring E. coli and total coliforms in natural spring water as related to recreational mountain areas. Environmental Monitoring and Assessmen 102: 131–137. https://doi.org/10.1007/s10661-005-4691-9

APHA (1998) Standard Methods for the Examination of Water and Wastewater, American Public Health Association, American Water Works Association and Water Environmental Federation, Washington DC.

Bauer-Gottwein P, Gondwe BRN, Charvet G, Marín LE, Rebolledo-Vieyra M, Merediz-Alonso G (2011) Review: The Yucatán Peninsula karst aquifer, Mexico. Hydrogeology Journal 19: 507–524. https://doi.org/10.1007/s10040-010-0699-5

Bojarczuk A, Jelonkiewicz Ł, Lenart-Boroń A (2018) The effect of anthropogenic and natural factors on the prevalence of physicochemical parameters of water and bacterial water quality indicators along the river Białka, southern Poland. Environmental Science and Pollution Research 25: 10102–10114. https://doi.org/10.1007/s11356-018-1212-2

Casanovas-Massana A, Blanch AR (2013) Characterization of microbial populations associated with natural swimming pools. International Journal of Hygiene and Environmental Health 216: 132–137. https://doi.org/10.1016/j.ijheh.2012.04.002

Derrien M, Árcega CF, Velázquez TN, Kantún MCA, Capella VS (2015) Sources and distribution of organic matter along the Ring of Cenotes, Yucatan, Mexico: Sterol markers and statistical approaches. Science of The Total Environmen 511: 223–229. https://doi.org/10.1016/j.scitotenv.2014.12.053

Escolero O, Marín LE, Steinch B, Pacheco JA, Molina-Maldonado A, Anzaldo JM (2005) Geochemistry of the hydrogeological reserve of Mérida, Yucatán, Mexico. Geofísica Internacional 44: 301–314.

Gómez-Nicolás M, Rebolledo-Vieyra M, Canto-Lugo E, Huerta-Quintanilla R, Ochoa-Sandoval P (2017) Connectivity in a Karst System using electrical resistivity tomography and network theory. Ground Water 56: 732–741. https://doi.org/10.1111/gwat.12618

Hijmans RJ, Williams E, Vennes C, Hijmans MRJ (2017) "Package Geosphere". https://cran.r-project.org/web/packages/geosphere/geosphere.pdf

Hoogesteijn-Reul AL, Febles-Patrón JL, Nava-Galindo VA (2015) La contaminación fecal en cenotes de interés turístico y recreacional del estado de Yucatán. Ingeniería 19–3: 169–175.

Instituto Nacional de Estadística y Geografía (INEGI) (2017) Anuario estadístico y geográfico de Quintana Roo 2017. México. https://www.inegi.org.mx/app/biblioteca/ficha.html?upc=702825087357

Kane K (2016) Impacts of Tourism on Water Quality in Quintana Roo, México. Master's Thesis. Northern Illinois University, Illinois.

Leal-Bautista RM, Lenczewski M, Acosta-González G, Grimaldo-Hernández C (2019) Evaluation of water quality through the distribution system in Cancún, Mexico. Sociedad y Ambiente: 53–75. https://doi.org/10.31840/sya.v0i21.2039

MacQueen J (1967) Some Methods for Classification and Analysis of Multivariate Observations. Proc. of the Fifth Berkeley Symposium on Math. Stat and Prob., vol. 1, 281–296.

McDonald AT, Chapman PJ, Fukasawa K (2008) The microbial status of natural waters in a protected wilderness area. Journal of Environmental Management 87: 600–608. https://doi.org/10.1016/j.jenvman.2007.10.007

Moore A, Lenczewski M, Leal-Bautista RM, Duvall M (2020) Groundwater microbial diversity and antibiotic resistance linked to human population density in Yucatan Peninsula, Mexico. Canadian Journal of Microbiology 66: 46–58. https://doi.org/10.1139/cjm-2019-0173

Mossel DAA (1982) Marker (index and indicator) organisms in food and drinking water. Semantics, ecology, taxonomy and enumeration. Antonie van Leeuwenhoek 48: 609–611. https://doi.org/10.1007/BF00399544

Nava-Galindo VA (2015) Percepción, conocimiento local y descripción de la calidad del agua de cenotes de interés turístico y recreacional. Master´s thesis. Centro de Investigación y Estudios Avanzados del Instituto Politécnico Nacional (CINVESTAV-IPN), Yucatán.

Norma Mexicana NMX-AA-042-SCFI-2015 (2015) Análisis de agua. Enumeración de organismos coliformes totales, organismos coliformes fecales (termotolerantes) y *Escherichia coli*. Método del número más probable en tubos múltiples. Secretaría de Economía, México.

Ogbondeminu FS, Gomwalk NE, Okuofu CE (1994) Faecal contamination of a tropical river in Nigeria and possible health risks. International Journal of Environmental Health Research 4: 147–155. https://doi.org/10.1080/09603129409356812

Pacheco AJ, Cabrera SA, Marín LE (2000) Bacteriological contamination in the karstic aquifer of Yucatán, Mexico. Geofísica Internacional 39: 285–291.

Ramos L, Vidal L, Vilardy S (2008) Analysis of the microbiological contamination (Total And Fecales coliforms) In The Bay Of Santa Marta, Colombian Caribbean. Acta Biológica Colombiana 13: 87–98.

Robusto C (1957) The Cosine-Haversine Formula. The American Mathematical Monthly 64: 38–40. https://doi.org/10.2307/2309088

Rochelle-Newall E, Nguyen TMH, Le TPQ, Sengtaheuanghoung O, Ribolzi O (2015) A short review of fecal indicator bacteria in tropical aquatic ecosystems: knowledge gaps and future directions. Frontiers in Microbiology 6: 1–15. https://doi.org/10.3389/fmicb.2015.00308

Rosiles-González G, Ávila-Torres G, Moreno-Valenzuela OA, Acosta-González G, Leal-Bautista RM, Grimaldo-Hernández CD, Brown JK, Chaidez-Quiroz C, Betancourt WQ, Gerba CP, Hernández-Zepeda C (2017) Occurrence of Pepper Mild Mottle Virus (PMMoV) in Groundwater from a Karst Aquifer System in the Yucatan Peninsula, Mexico. Food and Environmental Virology 9: 487–497. https://doi.org/10.1007/s12560-017-9309-1

Secretaria de Turismo (SECTUR) (2013) Agendas De Competitividad de los Destinos Turísticos de Turismo México, Estudio de Competitividad Turística del Destino Cancún, México. http://www.sectur.gob.mx/wp-content/uploads/2015/02/PDF-Cancun.pdf

Comisión Nacional del Agua (CONAGUA) (2015) Programa Hídrico Regional 2014-2018 de la región Hidrológico-Administrativa XII Península de Yucatán, México. https://www.gob.mx/cms/uploads/attachment/file/241045/PHR_09.06.16.compressed.pdf

Suárez-Morales E, Rivera-Arriaga E (1998) Hidrología y fauna acuática de los cenotes de la Península de Yucatán. Revista de la Sociedad Mexicana de Historia Natural 48: 37–47.

World Health Organization (WHO) (2004) Guidelines for Drinking Water Quality (3rd edn).

Williams E (2011) Aviation formulary V1.46. https://www.edwilliams.org/avform.htm