Enteroparasites in Preschool Children on the Pacific Region of Nicaragua

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Abstract. The aim of this study was to determine for the first time the prevalence of enteroparasites in preschool children originating from the seven departments of the Pacific region in Nicaragua. One stool sample of each of 1,217 children, from 6 months to 5 years of age, was collected and personal data were recorded on delivery of the container. Samples fixed in 10% formalin were processed by a formol-acetate concentration and a modified Ziehl-Neelsen technique. The overall prevalence of enteroparasite infections was 68.2% with a total of at least 20 species. *Blastocystis hominis* (45.5%), *Giardia intestinalis* (31.7%), *Trichuris trichiura* (8.2%), and *Ascaris lumbricoides* (5.2%) were the most prevalent protozoa and helminth species in the total study as well as in all departments. Protozoan prevalence presented a statistically significant difference by gender (male: 69.6%; female: 46.7%; $P < 0.001$), and males presented a higher *T. trichiura* infection rate than females (male: 9.9%; female 6.4%; $P < 0.035$). Protozoan prevalence increased with age with a statistically significant difference ($P < 0.001$). Helminths were always more prevalent in urban areas ($P < 0.0002$). Protozoan infections result statistically higher than helminth infections so that water-based transmission could be suspected. Based on the differences with respect to species and parasite prevalence between the seven departments in the Pacific region of Nicaragua, the exploration of local factors associated with the transmission of enteroparasites should also be considered to reduce infection.

INTRODUCTION

Although enteroparasitic infections are not life-threatening, they are harmful to children's health, impeding healthy growth, cognitive development, physical fitness, and the iron status, leading to malnutrition with the inherent complications and also affecting the immune response.^{1–3}

The prevalence of enteroparasites is a direct consequence of a set of constant factors, such as climate, food and water supplies, personal and community hygiene, sanitation, proximity to both domestic and wild animals, and socioeconomic conditions.⁴ Moreover, youth is an important factor for acquiring intestinal infections.^{4,5}

Worldwide, it is estimated that about 200 million preschool children are affected by restricted physical development,⁶ mainly in the tropical and subtropical regions of developing countries, such as sub-Saharan Africa, the Americas, China, and East Asia. In these regions, large-scale deworming programs are increasingly implemented in the preschool children population who is infected with at least one soil-transmitted helminth species.^{2,5,7–9}

The impact of intestinal parasites can either be clinically asymptomatic or have acute, respectively, chronic clinical symptoms.⁵ These symptoms are usually found in environments characterized by poverty, overcrowding, and insufficient sanitary facilities, for example, preschools of developing countries.^{10,11} Therefore, the study of such a young population is necessary to develop effective prevention and control strategies.¹²

Nicaragua, the largest country in the Central American isthmus, with approximately 6 million inhabitants, being bordered by Honduras to the north, Costa Rica to the south, the Pacific Ocean to the west and the Caribbean to the east, is the second poorest country among Latin America and Caribbean

(LAC) countries (Figure 1). Although several studies on intestinal parasites in older school children in Nicaragua have been already carried out,^{13–18} no previous work has so far investigated intestinal parasites in preschool children. The aim of this study was to determine the prevalence of enteroparasites in stool samples from these children originating from the departments of the Pacific region of Nicaragua, trying to evaluate which department, sex, age, and urban/rural background are most severely infected.

MATERIAL AND METHODS

Study area and population. Geographically, Nicaragua is divided into three regions: the Pacific region, having a population of 54% of the country, with 72.8% living in urban areas versus 27.2% living in rural areas; the Central and Northern regions, having a population of 32% (38.1% versus 61.9%); and the Caribbean region, with 14% of the total population (31.3% versus 68.7%). In general, urban and rural areas are normally not very different. However, rural areas are characterized by more dispersed houses without any infrastructures or sanitation facilities.

Between 2007 and 2011, a total of 1,217 children (591 females versus 626 males), from 6 months to 5 years of age (103 below the age of 1; 159 being 1 year old; 210 being 2 years old; 232 being 3 years old; 239 being 4 years old; and 274 being 5 years old), were randomly examined coming from urban (512) as well as rural (705) areas of the seven departments of the Pacific region of Nicaragua: Chinandega (112 preschools; 49 females versus 63 males; 13 below 1 year of age, 16 aged one, 21 aged two, 25 aged three, 16 aged four, and 21 aged five; and 59 urban versus 53 rural), León (141; 62 versus 79; 10, 24, 19, 31, 25 and 32; 43 versus 98), Managua (445; 233 versus 212; 28, 38, 70, 85, 102 and 122; 143 versus 302), Masaya (138; 66 versus 72; 12, 16, 29, 25, 28, and 28; 67 versus 71), Carazo (125; 58 versus 67; 13, 15, 18, 27, 23 and 29; 44 versus 81), Granada (138; 66 versus 72; 15, 30, 32, 17, 22 y 22; 94 versus 44) and Rivas (118; 57 versus 61; 12, 20, 21, 22, 23 y 20; 62 versus 56). Children were randomly selected from several

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FIGURE 1. Map of Nicaragua showing its departments and the seven departments surveyed comprising the Pacific region.

neighborhoods of similar characteristics. A minimum of 200 children was recruited each year along a 5-year study period. Concretely, 1,067 children were examined in their family homes in all the departments, whereas 150 children were analyzed at preschool establishments only in the Department of Managua.

Ethical approval. In all the departments concerned, meetings explaining the purpose of the study were held with the parents when visited at home, whereas the headteachers' permission was obtained at the different preschools. After signing a free informed consent form, agreeing on the children's participation, the parents and/or headteachers were instructed for the correct sample collection.

Stool samples. One stool sample per child was collected and personal data (name, sex, and age) were recorded on delivery of the container. In less than 24 hours, fecal specimens were fixed in 10% formalin and prepared for air transport to Spain. Two months later, parasitological observations were carried out at the Departamento de Parasitología (Valencia, Spain) with fixed samples processed by a formol-acetate concentration technique. The sediments of the concentration technique were used to obtain prevalence data. Based on this technique, the prevalences obtained for *Enterobius vermicularis* and *Strongyloides stercoralis* may not be considered definitive because anal swabs and the agar-plate method, respectively, would be the adequate techniques for the detection of pinworm eggs and geohelminth larvae. One aliquot of sediment obtained was stained using a modified Ziehl-Neelsen technique.¹⁹

Statistical analysis. Statistical analysis was performed using Open Epi (Open Source Epidemiologic Statistics for Public Health, Version 3.03a. www.OpenEpi.com). Differences between the results were compared with the χ^2 test, and all results were considered significant if $P < 0.05$. No further information (weight, height, environmental data, etc.) was available for other statistical studies.

RESULTS

Prevalence of enteroparasites. The entire enteroparasite spectrum in the Pacific region is made up of at least 12 protozoan species and at least eight helminth species (see Table 1). When analyzing the different departments, 12 species were found in Granada, the maximum number of protozoan species, whereas the departments of Chinandega, León, Carazo, and Rivas presented eight species. With respect to helminths, a total of six species were found in Managua, whereas Rivas and León presented two species, and three species were encountered in Masaya. The overall prevalence of enteroparasite infections was 68.2%, with protozoan infections being statistically higher than helminth infections (65.9% versus 13.1%; $P < 0.001$). The most prevalent protozoa species detected were *Blastocystis hominis* in the total study (45.5%) as well as in all departments (from 32.8% in Carazo to 69.6% in Chinandega), followed by *Giardia intestinalis* with a prevalence of 31.7% in the entire study, and variable differences between departments (see Table 1). León (81.6%) and Masaya (81.2%) presented the highest protozoan prevalence, whereas Managua (56.9%) and Carazo (65.6%) had the lowest prevalences.

Among helminths, *Trichuris trichiura* (8.2%) and *Ascaris lumbricoides* (5.2%) were the most prevalent species not only in the entire study but also in the departments studied (Table 1). The remaining helminth species appeared as isolated cases in various departments. The presence of *Hymenolepis nana* which appeared in all departments but Rivas was outstanding. The same goes for the prevalence of Ancylostomidae gen. sp. in Chinandega (3.6%). The departments with the highest helminth prevalences were Carazo, Masaya, and Chinandega, with 26.4%, 23.9%, and 21.4%, respectively.

Distribution of enteroparasites by gender. A statistically significant difference between the overall infection rates with protozoa based on gender was found (male: 69.6%; female: 46.7%; $P < 0.001$). Both males and females showed comparable infection rates with *B. hominis* (46.6% versus 43.8% respectively) and *G. intestinalis* (33.7% versus 29.6%), with no statistically significant differences. However, there was no statistically significant difference in the overall prevalence rate of helminth infection between males (14.7%) and females (11.2%), although a statistically significant ($P < 0.035$) difference was found for *T. trichiura*, with males presenting a higher infection rate than females (9.9% versus 6.4%, respectively) (Table 2).

Distribution of enteroparasites by age. Table 2 shows the overall infection rates among preschool children in the different age categories. The highest protozoan prevalence are reached as age increases, with statistically significant differences between the ages considered ($P < 0.001$). However, with respect to helminths, although prevalences increased with age, a statistically significant difference was only found in the case of *T. trichiura* ($P < 0.026$).

Distribution of enteroparasites by areas. Table 2 shows the detailed distribution of infection according to urban/rural areas. Protozoan parasites are always more prevalent in rural areas than in urban ones, yet without statistical significance. On the other hand, helminths are always more prevalent in urban areas, presenting statistically significant differences (Helminths: $P < 0.001$; *T. trichiura*: $P < 0.001$; *A. lumbricoides*: $P < 0.004$).

TABLE 1

Prevalence of intestinal parasitic infections among preschool children in each department surveyed and in the entire study of the Pacific region of Nicaragua

	Chinandega	Leon	Managua	Masaya	Carazo	Granada	Rivas	Total Pacific region	
	N = 112	N = 141	N = 445	N = 138	N = 125	N = 138	N = 118	N = 1,217	
	%	%	%	%	%	%	%	n	% (95% CI)
Protozoa	78.5	81.6	56	78.9	57.6	63.8	69.5	803	65.9 (63.3–68.6)
<i>Entamoeba coli</i>	23.2	32.6	18	30.4	16.8	15.2	17.8	257	21.1 (18.9–23.5)
<i>Entamoeba complex</i>	8	7.8	4.5	7.2	5.6	6.5	7.6	75	6.2 (4.9–7.6)
<i>Entamoeba hartmanni</i>	9.8	14.2	8.3	12.3	7.2	12.3	10.2	123	10.1 (8.5–11.9)
<i>Endolimax nana</i>	21.4	21.1	12.1	19.6	8	7.9	13.6	176	14.5 (12.6–16.5)
<i>Iodamoeba buetschlii</i>	1.8	0.7	2.7	5.1	1.6	3.6	2.5	32	2.6 (1.8–3.6)
<i>Giardia intestinalis</i>	35.7	30.5	30.3	37.7	28	37.7	24.6	386	31.7 (29.2–34.4)
<i>Chilomastix mensnili</i>	4.5	4.9	2.9	5.8	0	7.9	0.8	45	3.7 (2.7–4.9)
<i>Dientamoeba fragilis</i>	0	0	0	0	0	0.7	0	1	0.08 (0.004–0.4)
<i>Retortamonas intestinalis</i>	0	0	0.2	0.7	0.8	1.4	0	5	0.4 (0.2–0.9)
<i>Enteromonas hominis</i>	0	0	0	0	0	0.7	0	1	0.08 (0.004–0.4)
<i>Cryptosporidium</i> sp.	0	0	0.7	0	0	0.7	0	4	0.3 (0.1–0.8)
<i>Blastocystis hominis</i>	69.6	68.1	35.5	52.2	32.8	33.3	53.4	554	45.5 (42.7–48.3)
Helminths	21.4	5.7	6.7	23.9	26.4	18.1	5.1	159	13.1 (11.3–15.1)
<i>Hymenolepis nana</i>	1.8	0.7	1.1	3.6	1.6	5.8	0	23	1.9 (1.2–2.8)
<i>Hymenolepis diminuta</i>	0	0	0.2	0	0	0	0	1	0.08 (0.004–0.4)
<i>Enterobius vermicularis</i>	0	0	0	0	0	0.7	0	1	0.08 (0.004–0.4)
<i>Trichuris trichiura</i>	16.1	4.9	3.6	15.3	14.4	11.6	3.4	100	8.2 (6.8–9.9)
<i>Ascaris lumbricoides</i>	4.5	0	2.5	11.6	14.4	6.5	3.4	63	5.2 (4.0–6.5)
Ancylostomidae gen. sp.	3.6	0.7	0	0	0	0	0	5	0.4 (0.2–0.9)
<i>Strongyloides stercoralis</i>	0	0	0.2	0	0.8	0	0	2	0.2 (0.03–0.5)
<i>Trichostrongylus</i> sp.	0	0	0.2	0	0	0	0	1	0.08 (0.004–0.4)
Total infected	79.5	81.6	56.9	81.2	65.6	69.6	70.3	830	68.2 (65.5–70.8)

CI = confidence interval.

The distribution of enteroparasites by polyparasitism.

Regarding polyparasitism, our study found preschool children in the Pacific region to be infected with more than one species (42.6% poly- versus 24.9% monoparasitism), which is reflected in the entire study as well as in individual departments in such a manner that prevalence decrease as the

number of species increases (Table 3). The detection of two preschool children, both 4-year old boys, harboring at least eight different species, one from Masaya (*Entamoeba histolytica*/*Entamoeba dispar*/*Entamoeba moshkovskii*, *Entamoeba coli*, *Entamoeba hartmanni*, *Endolimax nana*, *G. intestinalis*, *Chilomastix mensnili*, *B. hominis* and *T. trichiura*) and one from Rivas

TABLE 2

Distribution of the most prevalent protozoan and helminth infections among preschool children in the Pacific region of Nicaragua according to gender, age, and urban/rural areas

	Protozoa	<i>Blastocystis hominis</i>	<i>Giardia intestinalis</i>	Helminths	<i>Trichuris trichiura</i>	<i>Ascaris lumbricoides</i>
Sex	%	%	%	%	%	%
Boys	95% CI 69.6 65.9–73.2	95% CI 46.6 42.8–50.6	95% CI 33.7 30–37.5	95% CI 14.7 12.1–17.6	95% CI 9.9 7.7–12.4	95% CI 5.9 4.3–7.9
Girls	46.7 42.7–50.7	43.8 39.8–47.8	29.6 26–33.4	11.2 8.8–13.9	6.4 4.6–8.6	4.1 2.7–5.9
P value	< 0.001	0.352	0.140	0.080	0.035	0.178
Age category (year)						
< 1	30 21.8–39.5	21.4 14.3–30.1	7.8 3.7–14.2	5.8 2.4–11.7	1.9 0.3–6.3	1.9 0.3–6.3
1	55.3 47.6–62.9	33.3 26.3–40.9	27.6 21.1–35	8.8 4.9–14.3	5 2.4–9.3	3.1 1.2–6.8
2	58.6 51.8–65.1	39 32.6–45.8	33.3 27.2–39.9	12.8 8.8–17.9	7.6 4.6–11.8	6.7 3.8–10.7
3	68.9 62.8–74.7	50 43.6–56.4	34.9 28.9–41.2	12.9 9.1–17.7	7.3 4.5–11.2	3.4 1.6–6.4
4	80.3 74.9–85	58.9 52.7–65.1	36.4 30.5–42.7	15.9 11.7–20.9	11.3 7.7–15.8	5.8 3.4–9.4
5	77.3 72.1–82	52.2 46.3–58.1	34.3 28.9–40.1	16.1 12.1–20.8	10.6 7.3–14.7	5.8 3.5–9.1
P value	< 0.001	< 0.001	< 0.001	0.056	0.026	0.274
Areas						
Urban	64.6 60.4–68.7	41.9 37.8–46.3	32 28.1–36.2	17.9 14.8–21.5	11.3 8.8–14.3	7.6 5.5–10.2
Rural	68.4 64.9–71.7	45.5 41.8–49.2	31.9 28.6–35.4	10.4 8.3–12.8	6.1 4.5–8.1	3.7 2.5–5.3
P value	0.159	0.217	0.956	0.0002	0.0017	0.004

CI = confidence interval. Bold values indicate significant results.

TABLE 3

Percentage (%) of mono- and polyparasitism species in the entire study of the Pacific region of Nicaragua, as well as in the seven departments surveyed

	Total Pacific region	Chinandega	León	Managua	Masaya	Carazo	Granada	Rivas
	% (95% CI)	% (95% CI)	% (95% CI)	% (95% CI)	% (95% CI)	% (95% CI)	% (95% CI)	% (95% CI)
Monoparasitism	24.9 (22.5–27.4)	22.4 (15.3–30.7)	22.9 (15.7–29.4)	21.1 17.7–25.3	28.3 21.2–36.2	26.4 19.2–34.6	25.4 19.9–34.7	36.4 28.1–45.4
Polyparasitism	42.6 39.9–45.4	57.1 47.8–66.1	59.0 51.3–67.4	33.7 29.2–37.9	52.9 44.5–61.1	39.2 30.9–47.9	44.2 36.1–52.6	33.1 25–41.9
2	19.3 17.2–21.7	19.6 13.1–27.8	29.1 22–36.9	17.1 13.8–21.1	20.3 14.2–27.6	20 13.7–27.7	18.8 12.9–26	16.2 10.3–23.6
3	11.9 10.3–13.9	19.6 13.1–27.8	19.1 13.3–26.3	8.8 6.4–11.9	10.1 5.9–10.1	12 7.1–18.6	15.2 9.9–21.9	6.8 3.2–12.5
4	6.3 5.1–7.8	11.6 6.6–18.6	5.7 2.7–10.5	4.3 2.7–6.7	12.3 7.6–18.6	4.8 1.9–9.7	4.3 1.8–8.8	6.8 3.2–12.5
5	2.9 2.1–4	4.5 1.6–9.6	305 1.3–7.7	2 1–3.9	6.5 3.2–11.6	2.4 0.6–6.4	2.2 0.6–5.8	1.7 0.3–5.5
6	1.4 0.8–2.2	1.8 0.3–5.8	1.4 0.2–4.6	1.3 0.5–3.1	2.9 0.9–6.8	–	–	–
7	0.4 0.2–0.9	–	0.7 0–3.4	0.2 0–1.4	–	–	1.4 0.2–4.7	0.8 0–4.1
8	0.2 0–0.5	–	–	–	0.7 0–3.5	–	–	0.8 0–4.1

CI = confidence interval.

(*E. histolytica*/*E. dispar*/*E. moshkovskii*, *E. coli*, *E. harmani*, *Iodamoeba buetschlii*, *G. intestinalis*, *B. hominis*, *T. trichiura* and *A. lumbricoides*) are illustrating cases.

DISCUSSION

This is the first time that intestinal parasitic infections among preschool children from the Central American isthmus have been studied. It is interesting to note that previous studies on preschool children are mainly from other continents.^{5,12,20} The main objective of epidemiological studies on the prevalence of intestinal parasitic infections is to contribute to effective prevention and control strategies, as is the case of the present study in the different departments of Nicaragua.

The parasite spectrum detected in Nicaraguan preschool children, with a total of at least 20 species, constitutes a parasitic framework larger than that known for schoolchildren in Nicaragua to date.^{13,14,16–18} The departments presented differences with respect to species, even with regard to parasite prevalences, likely to be due to advances in public health, as well as measures within the national deworming program against soil-transmitted helminthes.^{21,22}

According to the results obtained, *B. hominis* was the most prevalent intestinal species (45.5%), which is in line with a study conducted in schoolchildren from the Nicaraguan Corn Island archipelago (41%), although *Blastocystis* with *Isospora* was diagnosed in that study.¹³ Nevertheless, in the present study, the prevalence was lower compared with the two school-based studies carried out in the other two Caribbean departments of Nicaragua (always with a prevalence above 50%).^{17,18} In other Caribbean countries, *B. hominis* prevalence in schoolchildren are considerably lower.^{23–25}

In the entire study, *G. intestinalis* with a prevalence of 31.7% was the second most prevalent intestinal species in preschool children. This result is comparable to that encountered in school children from the Department of Río San Juan (32.5%),¹⁷ but being in contrast with studies carried out in school children originating from other Nicaraguan departments, always with prevalence below 30%.^{13–15,18} Although

the study population differs, in other Caribbean countries, *G. intestinalis* shows higher prevalence than in our preschool study.^{26,27}

In our study, *T. trichiura* was the most prevalent intestinal helminth species (8.2%) in all departments, which is in agreement with other studies carried out in school children in Nicaragua,^{13,16–18} as well as in the Central American isthmus.^{28–33} These results contrast with the dominance of *A. lumbricoides* and the hookworm in other areas in the LAC region.^{14,34–39}

Our study revealed statistical differences regarding gender in relation to protozoa in the total study, even in helminths with *T. trichiura*, in boys. These differences could be attributed to the fact that boys are more exposed to sources of infection related to their playing habits and other indoor activities and hardly ever adopt hygienic practices. These results are consistent with some studies performed in preschool children in other countries,^{5,20,40} proposing a different sociobehavioral gender role in the area surveyed but also disagrees with other studies.^{11,28,41,42} Recently, a survey carried out in El Salvador also demonstrated that school-age boys present a higher prevalence of *T. trichiura* infections.³³

Prevalence increase with age with statistical differences in protozoan species as well as *T. trichiura*. These results are consistent with studies carried out in preschool children in various countries,^{12,34,43,44} while being in contrast with other reports.^{5,28} The increase in prevalence when the preschool age increases could be related to the child's personal autonomy. Children of 1 and 2 years of age are under surveillance most of the time and those from 3 to 5 years start schooling and have more autonomy, less surveillance, and fewer hygienic habits are exposed to more risk factors and, finally, are more often infected than younger ones.

The present study showed distinct situations in both areas, resulting particularly relevant, with statistically significant differences, helminths being more prevalent in urban areas. In this context, it is noteworthy that the Pacific region of Nicaragua is the most developed part of the country, where the highest percentage of the population is concentrated in urban

areas. In fact, many families live in precarious conditions, lacking basic infrastructures as well as services, affecting their quality of life in a negative way, leading to the appearance of shanty towns, and problems of helminth parasitoses are exacerbated, fundamentally in the lowest age group, also being the most vulnerable segment of the population.

All intestinal parasites detected in this study, with the exception of the *Hymenolepis diminuta* case encountered in Managua, are monoxenous, evidently transmitted through water, food, and soil contaminated with feces or indirectly through poor hygienic and living conditions. The fact that protozoan prevalence show a clear statistical significance with respect to helminths is indicative of water-based transmission, as preschool children have relatively limited contact with soil, and consequently with the infective stages of soil transmitted helminth species. Also, mass drug administration (MDA) campaigns in preschool children may have been successful as well.

Our data confirm that polyparasitism is likely to be the norm rather than the exception in tropical and subtropical areas^{12,18,45} and the increasing number of parasites harbored in any individual may result in an increase in morbidity indicators.⁴⁶ In this sense, it is interesting that the occurrence of different parasite species in the gastrointestinal tract might mean a predisposition for the infection with other enteropathogens, modulating the immunological response to other species and thus facilitate secondary infections or even different degrees of polyparasitism.^{47,48} In fact, the detection of several protozoan species of a commensal nature in feces, although without clinical relevance, is important from an epidemiological viewpoint, as it is indicative of a lack in sanitary-hygienic education.

Considering the age range studied, the prevalence results must be considered high, even more so when taking into account that several problems could not be avoided in this study, such as 1) stool samples were prepared once, whereas for standard diagnosis, at least three samples are necessary, which is likely to cause an underestimation of the prevalence of intestinal parasites because of the temporal variation in egg excretion and cyst shedding along hours or even days; 2) the diagnostic techniques used have a relatively low sensitivity for the detection of certain parasite species (e.g., *E. vermicularis*, *S. stercoralis*); 3) the Kato-Katz technique, recommended by the World Health Organization for epidemiological STH surveys was not applied,⁴⁹ making it impossible to detect the intensity of parasite infection, and thus the parasite burden in preschool children; and 4) the possible risk factors related to the parasite species encountered cannot be discussed as a consequence of the lack of additional data.

This study shows the large spectrum of species detected in preschool children of the Pacific region of Nicaragua, with a high protozoan prevalence that might pose a major public health concern related to almost unavoidable water-based infection. Based on the differences with respect to species and parasite prevalence among the seven departments in the Pacific region of Nicaragua, an exploration of local factors associated with the transmission of enteroparasites should also be considered to reduce infection. Consequently, the Nicaraguan government together with MDA programs should also consider strategies of intervention to develop safe access to water and sanitation and hygiene, aiming at the reduction of the prevalence of intestinal parasites in preschool children.

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