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OCCURRENCE OF NAEGLERIA AND ACANTHAMOEBA IN SURFACE WATER (GOMTI RIVER) SAMPLES FROM LUCKNOW CITY

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ABSTRACT Water is a basic need of human for multiple requirements like, drinking, washing, bathing, swimming, cooking and is used for various other purposes such as energy production, industrial operation, navigation, agriculture, aquaculture etc. The microbial pathogens present in water may be either of faecal or non-faecal origin. Among the organism of non-faecal origin certain protozoa cause serious human health disorders after the consumption of contamination water by both human and domestic animal. Amphizoic amoebae are the most common ubiquitous protozoan organisms occurring in almost every natural and artificial body of water and are known to cause fatal diseases in human being and domestic animals. Free-living amoebae (FLA) are belonging to genera Naegleria and Acanthamoeba which as opportunistic pathogen causes fatal human disease affecting central nervous system and eye. These amoebae cause fatal Primary Amoebic Encephalitis (PAM), Granulomatous Amoebic Encephalitis (GAE) and Amoebic Keratitis in human being. In Lucknow, large number of human population living near or around of rivers and uses such water for daily uses and they are at high risk for infection.

KEYWORDS : Naegleria, Acanthamoeba, Primary Amoebic Encephalitis (PAM), Granulomatous Amoebic Encephalitis (GAE) and Amoebic Keratitis (AK).

INTRODUCTION

Water contamination is one of the most serious natural issues. Water in river contaminated with many pathogenic microbes which can endanger for human health and life.

Water bodies are heavily contaminated with a number of microorganisms both free-living as well as pathogenic. Bacteria, protozoan, helminthes, nematodes, etc. find their way into water through various human activities, domestic and industrial wastes. A potential health risk exists due to the presence of microbial and viral enteric pathogen in water destined for human consumption. These microorganisms are responsible for various water borne diseases like diarrhea, gastroenteritis, cholera, typhoid, meningitis, amoebiasis and other diseases.

Amoebae are the most common ubiquitous protozoan organisms occurring in almost every natural and artificial body of water such as lakes, rivers, dams, wells, ponds, tanks, swimming pools etc. small free-living amoebae, mainly found in water and soil are known to cause fatal diseases in human being and domestic animals.

Discoveries made in the last four decade have established the existence of small group of free-living amoebae belonging to genera *Naegleria* and *Acanthamoeba* which as opportunistic pathogen cause fatal human disease affecting central nervous system and eye. These amoebae cause fatal Primary Amoebic Encephalitis (PAM), Granulomatous Amoebic Encephalitis (GAE) and Amoebic Keratitis in human being.

MATERIAL AND METHODS

In the capital of Uttar Pradesh, Lucknow, river Gomti is the main source of water. In Lucknow, the river Gomti water majorly contaminated by numbers of manmade activity like discharge of faecal matters and domestic sewage, washing of cloths etc. Samples for studies were collected from different ghates of the river which showed maximum human activity in and around the river. Five such areas viz. Kudia ghat, Daliganj ghat, Karounda ghat, Panchwati Ghat, and Pipra ghat were selected as sample sites for isolation of amoebae.

About two liters of water sample was collected from each water body and filtered through sterile filter paper (Whatman No.-1) in a conical funnel. Sediment was collected in the cone of filter paper. About one cm cone of filter paper was cut placed on the thick suspension of *Escherichia coli* and incubated in BOD incubator at 28^0C-37^0C for 10-12 days. The culture was then observe d under microscope for the growth of amoebae.

For the isolation of amoebae, 15-20 ml of sterilized non-nutrient agar (2.5%w/v) with 6.6-6.8 pH poured in to pre-sterilized petridish (Borosil 9-10 cm in diameter) and allowed to s et for 24 hours. *Escherichia coli* grown on the surface of nutrient agar slants (pH-7.2) was used as food for amobae. Bacterial culture was scraped with **nichrome** loop from the nutrient agar tube and spread as thick suspension on the solidified non-nutrient agar surface in the form circular patch or bacterial circle (2-3) of 22-25 mm / 1.5-2.0 cm in diameter.

RESULT AND DISCUSSION

The Gomti River is main source of water for people in Lucknow; many people use this water source directly and indirectly. Keeping this in mind water samples were collected from different sites of Gomti River Ghats.

The maximum numbers of amoebic strains of *Naegleria* and *Acanthamoeba* were isolated from surface water of Gomti River viz. *Acanthamoeba culbertsoni, Naegleria fowleri, Acanthamoeba polyphaga, Acanthamoeba rhysodes* and *Naegleria gruberi* (Table-1 and Plate-1-2).

Table-1 Strains Of Naegleria And Acanthamoeba Isolated From Surface Water Of Different Ghats Of Gomti River, Lucknow

S.No.	SampleSites	Strains	AmphizoicAmoebae
1	Kudia Ghat	G-1	*Acanthamoebaculbertsoni*
		G-2	*Naegleriafowleri*
2	Daliganj Ghat	G-3	*Acanthamoebapolyphaga*
		G-4	*Acanthamoebarhysodes*
		G-5	*Naegleriafowleri*
		G-6	*Naegleriagruberi*
3	Karounda Ghat	G-7	*Acanthamoebapolyphaga*
		G-8	*Naegleriafowleri*
4	ChatMela Ghat	G-9	*Naegleriafowleri*
		G-10	*Acanthamoebaculbertsoni*
5	Pipra Ghat	G-11	*Acanthamoebarhysodes*
		G-12	*Acanthamoebapolyphaga*
		G-13	*Naegleriagruberi*

PLATE-1

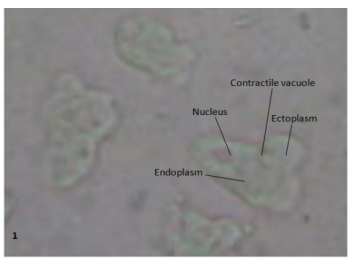

Fig-1 Show trophozoite of Naegleria sp.

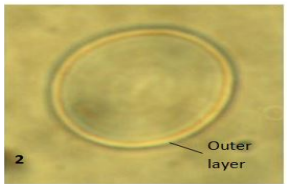

Fig-2 Show cyst of Naegleria gruberi-double layered

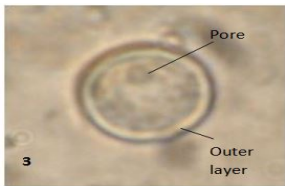

Fig-3 Show cyst of Naegleria fowleri, single layered with pore and outer gelatinous covering

PLATE-2

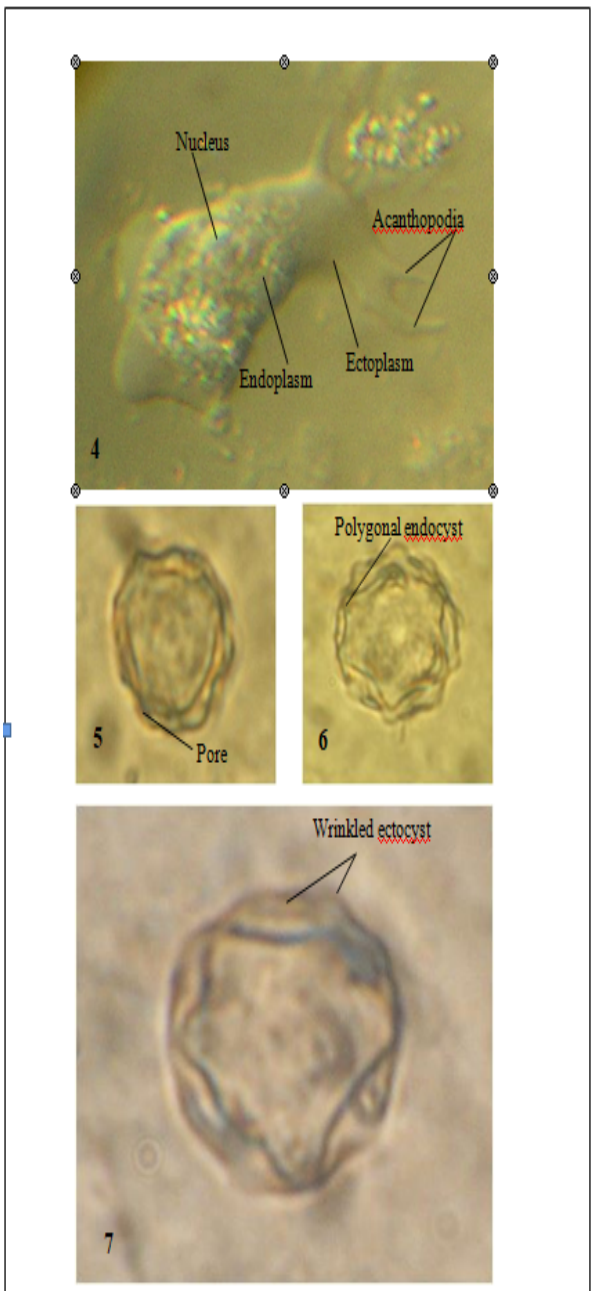

Fig-4 show trophozoite of *Acanthamoeba sp.*, acanthpodia
Fig-5 show cyst of *Acanthamoeba culbertsoni*, double layered with pore
Fig-6 show cyst of *Acanthamoeba polyphaga*, polyhedral and double layered
Fig-7 show cyst of *Acanthamoeba rhysodes*, double layered

Naegleria sp. isolated from surface water was *N. fowleri* (strain G-2, G-5, G-8 and G-9 from Kudia Ghat, Daliganj Ghat, Karounda Ghat and Chat Mela Ghat respectively, Plate -1) and *N. gruberi* (strain G-6 and G-13 from Daliganj Ghat and Pipra Ghat respectively Plate -1). Mansour *et al.* [16] has also reported that *Naegleria gruberi* was dominant in all surface water.

Acanthamoeba sp. isolated from surface water was *Acanthamoeba culbertsoni* (strain G-1 and G-10 from Kudia Ghat and Chat Mela Ghat, Plate -2); *Acanthamoeba polyphaga* (strain G-3; G-7 and G-12 from Daliganj Ghat, Karounda Ghat and Pipra Ghat respectively, Plate -2) and *Acanthamoeba rhysodes* (strain G-4 and G-11 from Daliganj Ghat and Pipra Ghat, Plate -2).

Most pathogenic strains of Amphizoic amoebae were isolated from river water (surface water) as compared to ground water in conformity with Hoffman and Michel [10]. The occurrence of pathogenic amoebae from different water sources which are used by humans for drinking and other purposes is an alarming situation. Pathogenic *Naegleria fowleri*, the causative agent of fatal PAM disease in a five month old infant from India [21] has been traced and isolated from a well. Pathogenic *Naegleria* spp. produce amoebo-flagellate stage in its life cycle, so chances of infection by *Naegleria* are maximum during swimming, diving and bathing in contaminated water of river and consequently may cause PAM [11]. The diseases Primary Amoebic Meningoencephalitis (PAM), Granulomatous Amoebic Encephalitis (GAE) are life threatening diseases whereas Amoebic Keratitis results in eventual lose of eye.

The Amphizoic amoebae have been isolated from various substrate, such as surface water sources like lakes, rivers, summer resorts, thermally polluted discharges [7], from artificially heated water [8], [7], from underground water like artificial well [19], from domestic tap water [4], [17], [13], from sewage and soil [20].

Kong [14] reported *Acanthamoeba* spp. from tap water in Korea. Ahmad *et.al*. [2] reported presence of *Acanthamoeba* and *Naegleria* speceies from natural water resources in Lucknow. Tung *et al*. [22] isolated *N. Fowleri* from hot spring and *Acanthamoeba* and *Naegleria* spp. from recreational water in Taiwan. Lass *et.al*. [15] isolated *Acanthamoeba* spp. from water sources.

Armand *et.al.* [2] reported *Acanthamoeba* species from water supply. The presence of *Acanthamoeba* is quite large in environmental sources. This will make vulnerable to human for fatal disease infections like granulomatous encephalitis or amoebic keratitis during daily use of water like drinking and bathing for survival [1].

De Jonckheere [6] and CDCP [5] reported the presence of *N. fowleri* from freshwater resources like swimming pools, hot springs, rivers, lakes and ponds, during the summer months. Johnson [12] and Montalbano *et.al.* [18] detected the prevalence of *N. fowleri* in river water that used for swimming or in swimming pool in Italy, New Zealand and USA. [9], [19].

CONCLUSION

The study focuses on two highly pathogenic genera of Amphizoic amoebae viz. *Naegleria* and *Acanthamoeba* because they are responsible for fetal diseases in human. Therefore the sample site was selected where the human activity has been reported maximum through different Ghats of Gomti Rivers especially in summer seasons. The result of present study confirmed that *Naegleria* and *Acanthamoeba* species colonized almost every Ghats. Surface water (Gomti River) is used by a large number of people on daily basis for many day to day requirements. The activity increases during summer months and during certain religious festivals especially around the different Ghats (sample site areas). The maximum samples showed presence of pathogenically active species of *Naegleria* and *Acanthamoeba* during summer months of years on every Ghats of Gomti River. These species, especially the fatal *N. fowleri* is more abundant in Kudia Ghat, Daliganj Ghat, Karounda Ghat and Chat Mela Ghat of Gomti Riverss. Whereas *Acanthamoeba polyphaga* colonized in water of Daliganj Ghat, Karounda Ghat and Pipra Ghat respectively. Isolation of these amoebae from surface water is an indicator of health hazard if infection occurs opportunistically or accidently during swimming, bathing and drinking the water by human and domestic animals causes serious disease like amoebae cause fatal Primary Amoebic Encephalitis (PAM), Granulomatous Amoebic Encephalitis (GAE) and amoebic keratitis in human or domestic animals.

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