



DEVELOPED ENDEMIC CONSORTIA USED IN UASB REACTOR TREATING MUNICIPAL WASTEWATER UNDER THE INFLUENCE OF pH/SALINITY AND HYDRAULIC RETENTION TIME

Kanhaiya Kumar Singh^{1*}, Vaishya R. C¹ and Arnav Gupta²

¹Department of Civil Engineering, Motilal Nehru National Institute of Technology-Allahabad, 211004, U.P., India

²Department of Civil Engineering, Birla Institute of Technology & Science, Pilani-333031, Rajasthan, India

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ABSTRACT

The upflow anaerobic sludge blanket reactor seeded with endemic consortia created from municipal wastewater isolates was utilized to treat raw domestic wastewater of medium strength. The UASB reactor required a time of 90 days to start up. The execution of reactor to treat municipal wastewater was upgraded with an expansion in the sludge age, pH/Salinity and Hydraulic Retention Time. At early sludge age (60 days), chemical oxygen demand (COD) and biochemical oxygen demand (BOD) expulsion by the reactor was in the scope of 59- 71%. Be that as it may, chemical oxygen demand (COD) and biochemical oxygen demand (BOD) evacuation proficiency of the reactors hoisted to the scope of 79-81% at sludge age of 150 days. To put it plainly, general execution of reactor was ideal at sludge age extending from 120 to 150 days, pH 7 and salinity 1%(w/v). At Hydraulic Retention Time (HRT) of 7.5 hrs the chemical oxygen demand (COD), suspended solids (TSS) and sulfate evacuation proficiency of UASB reactor achieved 81%, 75% and 76%, separately. The rate of evacuation of these parameters be that as it may, bit by bit declined with expanding Hydraulic Retention Time. The UASB innovation gives a minimal effort framework to the immediate treatment of municipal wastewater and can be connected in smallfraternity.

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INTRODUCTION

Among the different treatment innovations, anaerobic treatment frameworks are being empowered on account of a few focal points, including low development costs, little land necessities, low overabundance muck creation, plain activity and upkeep, vitality age as biogas (1) and strength regarding COD evacuation productivity (2), pH steadiness and recuperation time (3). Various analysts have prescribed anaerobic innovation like UASB reactor for the treatment of sewage in tropical and subtropical areas (4). The start-up of UASB reactors is a confounded procedure and various components, including wastewater qualities, acclimatization of seed slime, pH, supplements, nearness of harmful mixes, stacking rate, upflowvelocity (Vup), Hydraulic Retention Time (HRT), fluid blending and reactor configuration influence the development of sludge bed (5). The pH significantly impacts the development and survival of microorganisms. Albeit anaerobic treatment is conceivable at restricted pH and saltiness reaches, low and high pH and saltiness fixation normally prompts a decrease in the most extreme particular development rate and methanogenic action (6). Methanogenic activity at low and high scope of pH and saltiness is 10- 20 times lower than the action at medium

range, which requires an expansion in the biomass in the reactor (10- 20 times) or to work at higher sludge retention time (SRT) and Hydraulic Retention Time (HRT) so as to accomplish a similar COD evacuation proficiency (7). Hydraulic Retention Time (HRT) is a standout amongst the most critical parameters influencing the execution of a UASB reactor when utilized for the treatment of metropolitan wastewater (8). The upflowvelocity is straightforwardly related with HRT and assumes an imperative part to ensnare suspended solids. Abatement in the upflowvelocity involves an expansion in HRT, which helps suspended solids' (SS) evacuation effectiveness of the framework (9). The COD evacuation productivity of a UASB reactor likewise diminishes at hoisted upflowvelocity on the grounds that higher upflowvelocity decreases the contact time amongst muck and wastewater notwithstanding crushing of sludge granules, and resultantly higher washout of solids (10). Be that as it may, a few researchers detailed no particular impact of HRT on the treatment productivity of UASB reactor (11). The distinction of supposition in academic group is might be because of the distinction in the reactor configuration, working methodology and scope of HRT. In the present examination the development of sludge bed in UASB reactors at first seeded with endemic consortia created from municipal wastewater detaches was utilized to treat municipal wastewater of medium quality. The impact of process conditions (HRT,

**Corresponding author: Kanhaiya Kumar Singh*

Department of Civil Engineering, Motilal Nehru National Institute of Technology-Allahabad, 211004, U.P., India

sludge age, pH and salinity) on the execution of reactors was then analyzed.

MATERIALS AND METHODS

Sample Collection and Identification of Bacterial Strains

Municipal Wastewater sample was collected from railway pulliya MNNIT Campus Allahabad district, India, in pre-uncontaminated bottle as reported by approved procedures from American Public Health Association and reassigned immediately to the testing room. Wastewater sample was gradually adulterated and inoculated on the mineral salt agar medium distinctly. Morphologically contrasting colonies were deserted and preserved at 4°C on mineral salt agar slants. The deodorized strains were identified by morphological and biochemical characteristics based on Bergey's Manual of Determinative Bacteriology (12).

Analytical methods

Alkalinity, pH, chemical oxygen demand (COD), biochemical oxygen demand (BOD), conductivity, turbidity, total dissolved solids (TDS), total suspended solids (TSS), total hardness, chlorides, sulfates, oil and grease, color SVI, specific gravity and VSS were analyzed according to the Standard Methods for the Examination of Water and Wastewater (13). Settling velocity of the granules was determined using column study. It was assumed that the granules settle as discrete particles during column study. Turbidity, conductivity and dissolved oxygen (DO) were determined by the metric method according to standard methods (13). Standard Methods (4500-SO42 E, Turbidimetric method), (4500-Cl C Titrimetric method), (5220 B Open reflux method) and (5210 B) were used for the determination of Sulfate, Chloride, Chemical oxygen demand (COD) and Biochemical oxygen demand (BOD), respectively. Standard Methods (4500-N C), (4500-P Vanadomolybdophosphoric acid colorimetric method) and (5310 B) were used for the determination of Total Nitrogen, Total Phosphorous and Total Organic Carbon (TOC), respectively. Standard Methods (2540 B to E) were applied for the determination of Total Solids, Total Dissolved solids (TDS), Total Suspended solids (TSS) and Volatile suspended solids. Standard Methods (5520 B Gravimetric method) and 2120 C were used for the determination of oil and grease contents and color, respectively.

Outline of UASB reactor configuration

A small scale anaerobic UASB (Upflow Anaerobic Sludge Blanket) reactor was utilized as a part of this examination. The setup comprised of a couple of UASB reactors, peristaltic pump, influent tank, gushing gathering tank and gas catching framework. The UASB reactor was made of Perspex material, including a tubular segment at the base and an extended area named as gas- liquid- strong separator (GLSS) at the best. Tubular segment was a 120 cm long segment with 7 cm interior distance across (ID) and a volume of 4.6 L. The length of the gas- liquid- strong separator was 40 cm and volume was 10.2 L. The GLSS segment was additionally separated into two sections; base half was decreased with a slant point (\emptyset) of 60 and best half was a 20 cm long segment with an interior distance across of 22 cm. A modified shelter was additionally joined with the best cover of GLSS keeping in mind the end goal to advance coagulation of suspended/colloidal particles, help the accumulation of suspended particles to upgrade the

gathering of biogas and to control the washout of particles (14).

RESULTS AND DISCUSSION

Identification of Bacterial Strains

Add up to 112 bacterial segregates were secluded on supplement agar medium. Out of these, 7 bacterial segregates indicated development on Wastewater Agar medium. The natural issue contained in the wastewater gives a substrate to these segregates. Each of the seven bacterial strains were distinguished through morphological and biochemical qualities and furthermore described at sub-atomic level by 16S ribosomal RNA quality investigations. These strains were distinguished as Bacillus Anthracis A9, Serratia Marcescens B7, Listeria Monocytogenes C3, Klebsiella Pneumonia D13, Streptococcus Pneumonia R5, Enterococcus Faecalis S2 and Staphylococcus Aureus T6. Diverse consortia arranged on the basis of permutation and combination though consortia 11(A9+B7+S2+T6) performed well for biodegradation of municipal sludge.

Sludge granulation and start-up of UASB reactor

To study about the impact of Consortia 11(A9+B7+S2+T6) alongside disinfected Cow Dung was nourished in the reactor for acclimatization of seed culture for bacterial development. The cow dung sludge contained transcendently organic matters. Total solids (TS) and volatile suspended solids (VSS) fixations in the seed slime were 50.2 and 31.5 g/L, separately. Amid the acclimatization of seed sludge, supplements (COD: Nitrogen: Phosphorus in proportion of 300:5:1) were provided to support sludge development by the expansion of Sucrose ($C_{12}H_{22}O_{11}$) and diammonium hydrogen phosphate ($(NH_4)_2HPO_4$). The Biochemical oxygen request (BOD)/ total organic carbon (TOC) proportion was 1 (13). The sludge in the UASB reactor was an inhomogeneous suspended mass amid initial three months. After that granulation began and sludge bed was balanced out in a time of 4 months and the nature of sludge was tantamount with the well develop sludge of a digester.

Table 1 Structure of municipal sewage

Parameters	Values
pH	7.32±0.24
COD(mg/l)	472.23±26.51
BOD ₅	232.53±22.51
Conductivity (mS/cm)	1.23±0.43
Turbidity(FTU)	68.28±6.94
TSS(mg/l)	378.21±39.93
Chlorides (mg/l)	59.89±9.87
Color(absorbance)	0.052±0.02

The shade of seed sludge changed from light dim to dull dark in a time of 1 month, which exhibited the start of seed sludge adjustment. The sterilized sewage wastewater (Table-1) was then acquainted with the reactor to adjust with sludge. The sludge bed was just a suspended biomass up to a time of sixty days. After that granulation of bio-solids ended up unmistakable, this demonstrated fruitful start-up of the reactor. Be that as it may, sludge granulation completely showed up after 70 days (figure 1).

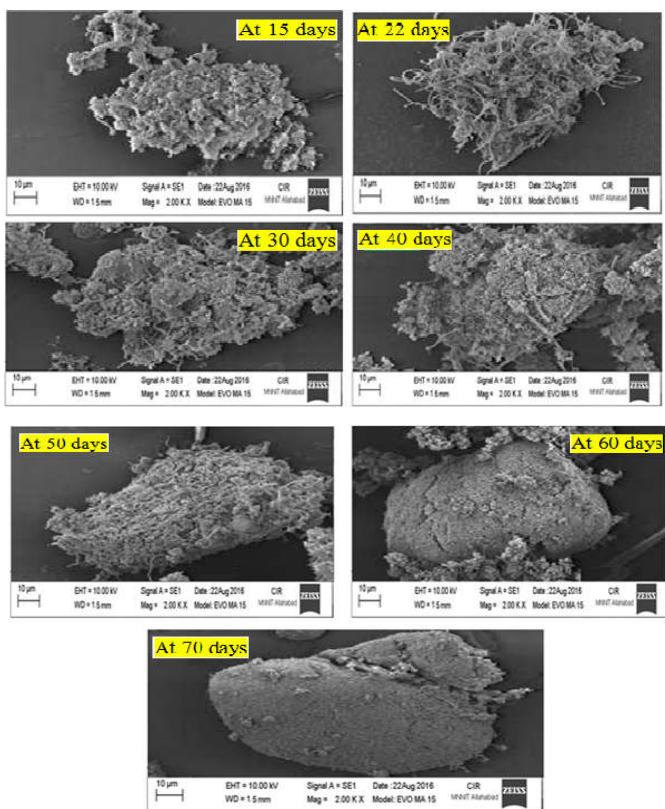


Fig 1 Scanning electron microscopic view of sludge granules at different day's from 15 to 70days

These outcomes are as per the discoveries announced in the writing. For example, LouweKooijmans and van Velsen (1986) detailed that start-up of UASB reactor seeded with processed cow dung manure excrement compost for household wastewater treatment at 25 °C required a time of a half year. Another investigation Vijayaraghavanand Ramanujan (1999) depicted that muck in anaerobic contact channel seeded with dairy animals excrement slurry was balanced out in a time of 160 days. Yasar (2006) immunized UASB reactor with actuated muck of journal wastewater treatment plants, and revealed 78 days start-up time for the reactor. Likewise a time of 147 days was required by Rajakumar *et al.* (2011) to begin a UASB channel. Figure 1 demonstrates UASB reactor profile amid startup and granulation.



Fig 2 UASB reactor profile during start-up and granulation

The start-up of UASB reactors is a muddled procedure and various components, including wastewater qualities, acclimatization of seed sludge, pH, supplements, nearness of toxic compounds, stacking rate, upflowvelocity, hydraulic retention time, fluid blending and reactor configuration influence the development of sludge bed (15).

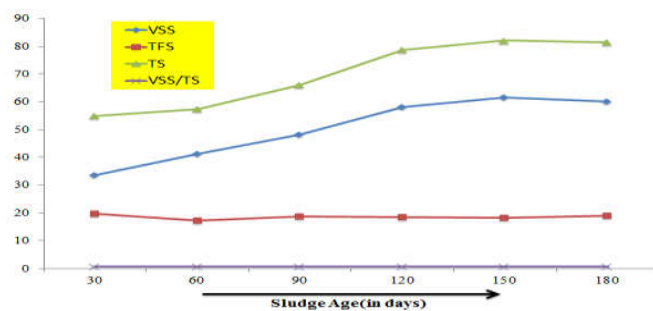


Fig 3 Sludge composition with respect to sludge age in UASB reactor

Variety in era required for the adjustment of the sludge may owe to a few elements, incorporating dissimilarities in wastewater piece, seed sludge, and type of reactor, sludge temperature, supplement substance, and sludge pH (16). Figure 3 demonstrates the impact of age on the sludgesynthesis as far as volatile suspended solids (VSS), add up to solids (TS), Total settled solids (TFS) substance in UASB reactor. The TS and VSS substance were 55.2 and 34.5 g/L, individually at the sludge age of 30 days. There was a general increment in TS and VSS substance as the sludge age expanded. The expansion in TS and VSS substance proceeded up to sludge age of 150 days, and TS and VSS substance were lifted to 80.5 and 62.3 g/L, individually for UASB reactor. A noteworthy increment in the VSS fixation when contrasted with TS was unmistakably a sign of the dynamic biomass development in the reactors as over 90% of VSS substance are because of dynamic biomass, and staying 10% are credited to non-biodegradable unpredictable solids and dead cell flotsam and jetsam (17). At sludge of 180 days, a decrease in TS and VSS substance of sludge in the two reactors showed up, which could be because of the liquidation of granules. The VSS/TS proportion is vital in deciding the sludge attributes and reflects biomass development and its quality. The VSS/TS proportion in UASB reactor step by step expanded (from 0.63 to 0.77) up to a sludge age of 150 days took after by a drop in this proportion at sludge age of 180 days. The distinction in VSS/TSS proportion may owe to a distinction in the substance and settleability of processed cow compost and enacted sludge (18).

Influence of pH

The impact of pH on the execution of a UASB reactor is critical on the grounds that it influences altogether the hydrolysis procedure, substrate use rate, settling of solids and gas exchange rates (19). The rate of anaerobic processing is quickly diminished as the pH of sludge bed is dipped under the pH range (8-4) (20). The ideal pH extend for the acidogenic stage can be affected by the attributes of the bolster and working conditions. Figure 4 demonstrates the expulsion productivity of all parameters on four distinctive pH (6.5, 7.0, 7.5 and 8.0) and consistent HRT and saltiness. Results from this and past investigations demonstrate that the runs (without pH control) related with high VFA creation were to a great extent worked at pH esteems in the vicinity of 5.0 and 5.3, which can be considered as an 'ideal' a similar pH go has been acquired in a UASB reactor treating an engineered slop(21). It has been as often as possible detailed that corrosive stage bioreactors were effectively worked at pH esteems in the vicinity of 5.0 and 6.0(22).

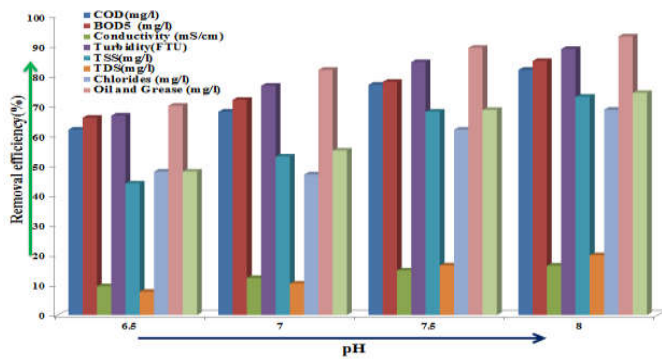


Fig 4 The performance of UASB reactors with different pH at constant 7.5 HRT and 1% (w/v) Salinity.

Acetic acid is the most prevalent product, as it is formed directly from the fermentation of carbohydrates and proteins, as well as during the anaerobic oxidation of lipids via a number of metabolic pathways (23). Propionic acid is formed primarily from carbohydrates, but it can also be produced in the digestion of the other two organic classes (24). On the other hand, butyric acid is mainly generated in the digestion of proteins and lipids. It may also be formed in the fermentation of carbohydrates from pyruvate via an alternative pathway (25). In mixed-culture fermentations, however, this pathway is considered as a rather minor one (26). The COD, BOD, and TSS removal efficiency of UASB reached 68%, 72% and 53%, respectively at room temperature and sludge age of 90 days. The improvement in the efficiency may owe to an increase in the digestion rate due to favorable temperature and relatively better developed sludge bed (27). These results also agree well with the findings of Lew *et al.* (2003), who reported 4% increase in the COD removal efficiency of a UASB reactor employed to treat domestic wastewater when the sludge temperature was increased from 14 to 20⁰ C. These findings are also in agreement with the results of other research workers (28) who reported COD and TSS removal efficiency of a UASB reactor inoculated with digested cow manure up to 78% and 75%, respectively at an operational temperature of 25⁰ C. Slightly lower TSS removal efficiency of the UASB reactors used in this study may owe to relatively less volume of sludge bed, which is insufficient to entrap non-settleable suspended solids (29). At sludge age of 150 days and 30⁰ C temperature, the UASB reactor showed COD, BOD, TSS and oil and grease removal up to 81%, 83%, 73% and 93%, respectively. The better performance of the reactors at this stage may be attributed to favorable sludge pH, well developed sludge granulation and increased growth of biomass which ultimately resulted in accelerated degradation of organic matter and entrapment of suspended solids (30). At sludge age of 180 days, the UASB reactor showed COD, BOD, and TSS removal of up to 82%, 85% and 73%, respectively. It was evident from the results that the performance of reactor was optimal at sludge age ranging from 120 to 150 days. Beyond sludge age of 150 days, the removal of pollution parameters by the reactors was marginal though the sludge pH was favorable for anaerobic digestion. The decline in efficiency may be due to liquidation of sludge, disengagement of entrapped solids and/or reduced rate of hydrolysis because enzymes involved in the hydrolysis are very sensitive to temperature (31).

Acidic corrosive is the most common item, as it is shaped specifically from the maturation of starches and proteins, and in addition amid the anaerobic oxidation of lipids by means of various metabolic pathways (32). Propionic corrosive is

shaped fundamentally from starches, however it can likewise be delivered in the assimilation of the other two natural classes (33). On the other hand, butyric corrosive is predominantly created in the absorption of proteins and lipids. It might likewise be shaped in the aging of starches from pyruvate by means of an elective pathway (34). In blended culture maturations, be that as it may, this pathway is considered as a somewhat minor one (35). The COD, BOD, and TSS evacuation productivity of UASB achieved 68%, 72% and 53%, separately at room temperature and slop age of 90 days. The change in the proficiency may owe to an expansion in the absorption rate because of great temperature and moderately better created sludge bed (36). These outcomes additionally concur well with the discoveries of Lew *et al.* (2003), who announced 4% expansion in the COD evacuation proficiency of a UASB reactor utilized to treat residential wastewater when the slop temperature was expanded from 14 to 20⁰ C. These discoveries are additionally in concurrence with the after effects of other research specialists who announced COD and TSS evacuation productivity of a UASB reactor immunized with processed dairy animals compost up to 78% and 75%, separately at an operational temperature of 25 C. Somewhat lower TSS expulsion productivity of the UASB reactors utilized as a part of this examination may owe to generally less volume of slime bed, which is deficient to capture non-settleable suspended solids (37). At sludge age of 150 days and 30 C temperature, the UASB reactor demonstrated COD, BOD, TSS and oil and oil evacuation up to 81%, 83%, 73% and 93%, individually. The better execution of the reactors at this stage might be credited to ideal muck pH, all around created sludge granulation and expanded development of biomass which at last brought about quickened corruption of natural issue and ensnarement of suspended solids (38). At sludge age of 180 days, the UASB reactor demonstrated COD, BOD, and TSS evacuation of up to 82%, 85% and 73%, individually. It was apparent from the outcomes that the execution of reactor was ideal at sludge age running from 120 to 150 days. Past sludge age of 150 days, the evacuation of contamination parameters by the reactors was peripheral however the slime pH was great for anaerobic absorption. The decrease in effectiveness might be because of liquidation of sludge, withdrawal of entangled solids or potentially lessened rate of hydrolysis since compounds associated with the hydrolysis are extremely delicate to temperature (39).

Influence of hydraulic retention times (HRTs)

The hydraulic retention time (HRT) is straight forwardly identified with upflow velocity (V_{up}) of influent in a UASB reactor. Henceforth, a satisfactory V_{up} and likewise HRT gives adequate contact amongst sludge and wastewater, lessens the development of gas pockets, separates the biomass from gas and resultantly upgrades TSS evacuation effectiveness of the framework (40).

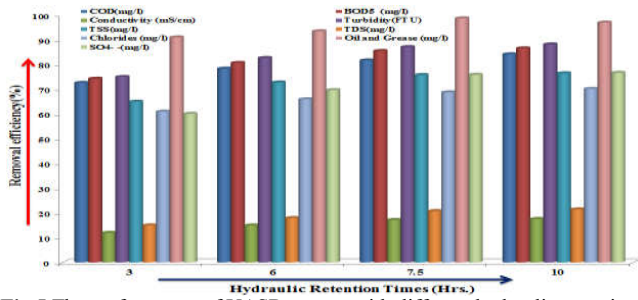


Fig 5 The performance of UASB reactor with different hydraulic retention times (HRTs) at constant pH7 and 1% (w/v) salinity.

Figure 5 demonstrates the impact of HRT on the execution of reactor at steady pH and saltiness. The outcomes uncovered that the COD, TSS and sulfate evacuation effectiveness of UASB reactor was 73%, 65% and 60%, individually at a pressure driven maintenance time of 3 h. The COD, TSS and sulfate evacuation proficiency of previous reactor was 84%, 77% and 76%, individually for later one at HRT of 10 h. Be that as it may, rate of evacuation of these parameters bit by bit declined with expanding water driven maintenance time. Comparable discoveries have likewise been accounted for in the writing (41). For example, Ruiz *et al.* (1998) announced that COD and TSS expulsion productivity of a lab scale UASB reactor treating household wastewater at 20 C was expanded from 53% to 73% and 63% to 80%, individually with an expansion in HRT from 4 to 8 h. Additionally, Nkemka and Murto (2010) revealed 81% COD expulsion effectiveness at HRT of 12 h in a UASB reactor treating kelp leachate and Zhang *et al.* (2012) detailed 92% COD evacuation effectiveness at HRT of 10 h in a UASB reactor treating sewage.

Influence of Salinity (w/v %)

Biodegradation of organics in such saline waste streams is accounted for to be poor because of dangerous impacts of sodium on the bacterial cells and Archaea causing cell lysis and demise. Medications getting salt had a tendency to have bring down quantities of microbes than the separate oil-altered control, however there was little consistency for this impact of salt on anaerobic muck microscopic organisms. Saltiness lessens the metabolic action of anaerobic slop microorganisms. Figure 6 demonstrates the impact of saltiness on the execution of reactor at consistent HRT and pH. The outcomes uncovered that the COD, TSS and sulfate expulsion productivity of UASB reactor was 71.4%, 64.8% and 61.7%, separately at 0.5% Saline focus (w/v). The COD, TSS and sulfate expulsion productivity of previous reactor was 54.2%, 56.5% and 46.7%, individually for later one at 2.0% Saline fixation (w/v).

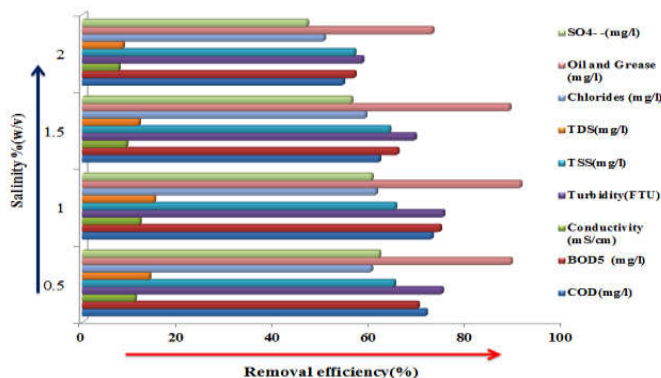


Fig 6 The performance of UASB reactor with different salinity at constant HRT 7.5 and pH 7.

Physical and substance treatment forms are vitality serious and more costly contrasted with organic procedures (42), subsequently the need to investigate the cutoff points of natural wastewater treatment in regards to saltiness and to extend these breaking points where conceivable. Due to a lower affectability of high-impact biomass to salt lethality contrasted with anaerobic biomass, most research is done on oxygen consuming treatment (43). Anaerobic organic wastewater treatment has a few focal points over oxygen consuming, for example, net vitality recuperation, lessened sludge creation and the likelihood to deal with higher volumetric stacking rates if arrangement of granular or immobilized biomass is conceivable. In the field of anaerobic wastewater treatment it is for the most part acknowledged that methanogenic movement is hindered above sodium levels of 5 g Na⁺/L (44). Notwithstanding, sodium lethality may rely upon the structure of wastewater because of conceivable hostile and synergistic impacts caused by different particles. In addition the kind of natural substrate for the most part decides the sort of microorganisms advancing in the reactors and consequently their affectability to sodium poisonous quality (45). Consequently, the capability of anaerobic treatment of saline wastewaters up to this point may have been to a great extent thought little of. On the off chance that high rate anaerobic bioreactors, for example, upflow anaerobic sludge blanket (UASB) for treatment of wastewater are connected, both microbial action and arrangement of granular microbial totals assumes a pivotal part on the procedure execution. After the underlying development of granules it is of equivalent significance to keep up a vigorous granular structure. Microbial granules of adequate quality and weight at that point permit the decoupling of water powered and slime maintenance times, accomplish high biomass fixations and allow the utilization of minimized reactors (46). The most ordinarily utilized techniques to improve the rate of granulation are expansion of multivalent cations, for example, calcium (150- 300 mg/L) (52) or potentially expansion of a help material, for example, powdered activated carbon (PAC) (47).

CONCLUSION

Following conclusions are drawn from this examination. The start-up of UASB reactor required a time of 90 days with created consortia. The execution of reactor to treat wastewater was improved with an expansion in the pH and sludge age. General execution of reactor was ideal at sludge age extending from 120 to 150 days, HRT 7.5, Salinity 0.5% (w/v) and pH shifting in the vicinity of 7 and 8. High saltiness went about as a specific weight on the predominant dynamic microbial and diminishing wealth of Methano-microbiales at hoisted salinity. The UASB innovation gives a minimal effort framework to the immediate treatment of municipal wastewater and can be connected in little groups where the wastewater stream variety is high because of stormy season or populace stack increments amid the traveler season or because of occasionally worked sustenance ventures.

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