Start-up of a decentralized pilot plant for the anaerobic treatment of domestic wastewater

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Abstract
The present study focuses on the set-up and start-up of a decentralised anaerobic pilot plant in order to produce domestic service water out of domestic wastewater. As there is no generally accepted definition for “service water” as with “drinking water”, it was defined as water that satisfies the requirements of the wastewater standard of Germany as well as the Bavarian bathing waters regulations. The pilot plant consists of a two-stage anaerobic digestion process (reactors 1 and 2) for the degradation of organic matter and a third-stage (reactor 3) for the removal of ammonium by means of the Anammox process. Each reactor was started up independently with synthetic wastewater having stage specific compositions. After the adaptation of the microorganisms to their new surroundings, a stepwise adjustment to municipal wastewater (MWW) occurred. The average removal efficiency of the two-stage anaerobic digestion process operated with 100 % MWW was about 62 % within 24 h of retention time. In the case of the anammox process, substrate limitation of nitrite led to a decline of removal efficiencies from 90 % at the beginning to 38 % at its minimum while adapting the microorganisms to municipal wastewater. After adjusting the NO2-N to NH4-N ratio to 1.14 the ammonium removal increased to a maximum of 95 %. After the start-up the degradation performance of the plant was tested for 200 days of operation. The two-stage anaerobic digestion had an average COD removal efficiency of 60 % and average outlet concentrations of 85 mg COD/l. In reactor 3 another 21 % of COD were removed until an average outlet COD of 39 mg/l was reached. Thus the self-defined service water limit value of 75 mg/l for COD was only achieved with the Anammox reactor in downstream. With an average outlet concentration of 1 mg NH4-N/l the self-defined service water limit value was also reached in the Anammox reactor. The average ammonium removal efficiency was about 96 %. Finally it can be concluded, that the start-up of the anaerobic wastewater treatment plant was successful.

Keywords
anaerobic organic digestion, Anammox process, service water, decentralised, start-up

INTRODUCTION
Realization of effective water cycles in households is an important aspect for saving water having drinking water quality. Not only treating but also recycling of domestic wastewater in order to reuse it as service water for minor domestic usage such as toilet flushing or cloths washing is a promising approach. Thus up to 60 % of fresh water can be replaced in households by service water (Herbst, 2008). In this context the necessity of decentralized systems that treat the incoming wastewater on site and thus don´t need long water transport distances inducing higher maintenance costs increases (Commission, 2005). The plant design of decentralized systems should be space efficient and odourless, as well as easy in use so that simple instructions are sufficient for operation (Massoud, et al., 2009; Herbst, 2008).

Due to low energy consumption, low sludge production, low space requirements and the production of useable energy in the form of biogas, anaerobic digestion is particularly attractive for organic removal in decentralized wastewater treatment systems (Batstone, 2006; Olsson, et al., 2013). Depending on the process design, meaning one-stage or two-stage operation, as well as the operational conditions, including temperature and pH, and the used reactor types, the removal
efficiencies for organic in the anaerobic digestion ranges in between 25% to 90%. Kobayashi et al. (1983) for example reached an organic removal efficiency of 73% for low strength domestic wastewater (288 mg COD/l) by using a lab-scale aerobic filter with a capacity of 11 ml/min. With an upflow anaerobic sludge blanket (UASB) reactor of 120 liters of volume Barbosa et al. (1989) achieved an average organic removal of 74% for treating raw domestic sewage (627 mg COD/l) within 4 h retention time. Domoso-Bravo et al. (2009) treated low-strength wastewater (500 mg COD/l) with a high fraction of particulate organic matter (70%, COD basis) by using a two-stage anaerobic system consisting of anaerobic sequencing batch reactors (ASBR). 69% and 50% of total COD removal efficiencies for organic loading rates (OLR) of 0.63 and 1.22 kgCOD/(m³*d) were achieved. Containing carbamite due to urine present in domestic wastewater, an additional treatment step has to be implemented subsequently to the anaerobic digestion in order to reduce the ammonia concentrations. In this context anaerobic ammonium oxidation (Anammox) represents an innovative possibility to oxidize ammonia to elementary nitrogen anaerobically by the use of nitrite (Ma et al., 2016; Strous et al., 1998; Van De Graaf et al., 1996). Fux et al. (2002) fed for example ammonium-rich wastewater of about 650 mg NH₄⁻/Nm³ in a semi-full scale sequencing batch reactor (SBR) with a capacity of 2 m³. Therefore nitrogen removal efficiencies of 85% to 99% and reduction rates in between 1.1-1.2 kgNH₄-N/(m³*d) were reached. Van der Star et al. (2007) could successfully start-up a full-scale reactor of 70 m³ treating 750 kgNH₄-N/d by using biomass of a 5 m³ enrichment reactor. Feeding synthetic wastewater similar to Van De Graaf et al. (1996) and setting 35°C as operation temperature, the enrichment reactor achieved conversion rates of about 5 kgNH₄-N/(m³*d). Nevertheless the practical application of the Anammox process ist still challenging. One big challenge is the slow growth rate of the microorganisms with a doubling time of 7 to 14 days causing slow start-up phases (Schmidt et al., 2003; Strous et al., 1998; Ali et al., 2015; Van der Star et al., 2007). Anaerobic digestion and anaerobic ammonium oxidation were mostly used for highly polluted wastewaters showing good degradation performances. Accordingly the adaptation of the microorganisms to low concentrations is one of the challenging steps for making anaerobic processes suitable for domestic wastewater treatment (Vega De Lille, 2015; Ali et al., 2015). The present study focuses the set-up and start-up of a decentralised anaerobic pilot plant combining the anaerobic digestion and Anammox-based process for the treatment of low strength domestic wastewater. Further aim of this project is to produce service water out of domestic wastewater that can be reused in households. In contrast to drinking quality water service water is not clearly defined or regulated. In the framework of the present project the generated service water follows the requirements of the German wastewater regulations (AbwV., 2016) and the Bavarian bathing waters regulations (BayBadeGewV, 2008). In addition the service water should be olfactory and aesthetic attractive. Allowed maximum concentrations for ammonia, total nitrogen (TN) and phosphor are 10 mg/l, 13 mg/l and 1 mg/l. The organic load represented by the sum parameters chemical oxygen demand (COD) should not overcome 75 mg/l. Being indicator microbes for feces polluted wastewater, E.coli and enterococci should be limited to concentrations of 900 cfu (colony-forming units)/100 ml und 330 cfu/100 ml (AbwV., 2016; BayBadeGewV, 2008).

MATERIAL AND METHODS

Set-up of the pilot plant
The pilot plant presented in this study consists of three biological stages wereof the first two stages, reactor 1 (R1) and reactor 2 (R2), are used for the anaerobic digestion of organic matter. The third-stage (reactor 3, R3) contains the Anammox based process in order to reduce the ammonium concentrations. Designed for a capacity of 2 m³/d the pilot plant is built up in the municipal sewage
plant of Erlangen, Germany (s. Figure 1). According to the decentralized purpose, a space efficient plant design was realized by integrating the process into two standard office containers that are connected to one room cell. With a total area of 28 m² and a ceiling height of 2.50 m the plant can also be easily installed in basements of apartment buildings.

Due to cost-effectiveness all three reactors with a capacity of 1200 liters each are designed as cylindrical tanks made out of polyethylene. Reactors 1 and 3 were designed as batch stirred tank reactors (BSTR) with a height of 1300 mm and a diameter of 1100 mm each, which corresponds to a filling height-diameter ratio of one. For suspending the microorganisms and homogenization of the reactor medium an axial flow field is induced by stirring in the BSTR. As a result the suspended bacteria in reactor 1 and 3 need to settle down before pumping the effluent to the next process step in order to guarantee biomass retention. For this reason the BSTRs are equipped with an outlet positioned at 200 mm of height from reactor bottom so that a retention volume of approximately 200 liters is provided.

Due to the symbiotic character of the acetogenic and methanogenic microorganisms a high space density for the bacteria in the reactor has to be ensured. Thus reactor 2 is built up as a fixed bed reactor (FBR) having 2100 mm in height and 900 mm in diameter. As support material completely three-dimensional permeable blocks of the type BIO-NET® from Norddeutsche Seekabelwerke GmbH are used. In the case of the FBR, the bacteria remain attached to the fixed bed and the down-stream recirculation through the fixed bed is essential for a good distribution.

As the microorganisms being used in the process are mesophilic, a temperature range in between 30°C and 40°C has to be realized (Grady, et al., 2011; Vega De Lille, 2015). Thus the wastewater has to be heated for good degradation performances. In this case a buffer tank of 1000 liters capacity equipped with a heating cartridge is used as pre-heating step. Furthermore the buffer tank facilitates the start-up phase of the three reactors and offers the possibility of buffering between the biological and post treatment. The tank is connected to the collector pipe after the pre-treatment basins of the communal sewage plant which remove big particles from the wastewater. Starting from the sampling point, one cubic meter per charge of fresh wastewater is pumped to the buffer tank in which it is heated up to the process operating temperature (35°C). Afterwards, the wastewater is transferred to the two-stage organic digestion where hydrolysis and acidogenesis are taking place in the first and acetogenesis and methanogenesis in the second reactor. Subsequently the wastewater is pumped to the third reactor (R3) where the Anammox process occurs. Finally the biologically treated wastewater is transferred to the post treatment consisting of two slow sand filter units and one activated carbon adsorber module in order to augment the service water quality by eliminating suspended solids, microorganisms and residual polluting loads like phosphate. The complete layout of the plant is shown in Figure 2.

Figure 1: Decentralised anaerobic pilot plant for producing domestic service water out of domestic wastewater. The plant is built up in the communal sewage plant of Erlangen, Germany. The plant is integrated in two standard office containers that are connected to one room cell with a total area of 28 m² and a ceiling height of 2.50 m.
Figure 2: Process and instrumentation scheme of the decentralised anaerobic pilot plant for producing domestic service water out of domestic wastewater.

Start-up and testing of the anaerobic pilot plant for the treatment of domestic wastewater
For start-up all reactors were operated separately at first in order to facilitate the adaptation of the microorganisms to their new surrounding conditions. Then R1 and R2 were interconnected initially by replacing stepwise the municipal wastewater (MWW) supplemented with additional organic acids by the outlet of R1. Finally R3 was linked to the anaerobic digestion by substituting the artificial wastewater by the outlet of R2.

Reactor 1 and 2 were inoculated each with 60 liters of seeding sludge coming from a two-stage anaerobic digester in Obermichelsbach (Germany) where sustainable vegetable products are degraded. Both reactors were fed right from the beginning with mechanically pretreated MWW. Degrading predominantly short chain organic acids in the acetogenic and methanogenic reaction phase, reactor 2 had to be fed with additional substrate as far as the connection to the hydrolysis and acidogenesis process step was implemented (phase I). As suggested by Kuba, et al. (1990) and Vega De Lille (2015) acetic acid, propionic acid and butyric acid were added in a ratio of 2:1:1 to reactor 2. As municipal wastewater was used from the beginning, trace elements were not needed. After the adaptation phase the interconnection of the two reactors was carried out stepwise (phase II). The inlet of R2 being 1000 liter of fresh MWW supplemented with the indicating organic acids at the beginning was replaced first by 30 %, then by 50 % and finally by 100 % of the outlet of reactor 1. After completing the interconnection of the two reactors no additional substrate was fed anymore (phase III).

Another parameter influencing the degradation performance of the microorganisms is the pH-value. Studies in lab-scale showed that adjusting the pH according to the microorganisms’ optimum has an influence on the intermediate products in a way that the degradation performance is being improved. In hydrolysis and acidogenesis step the optimum pH is in between 4.5 and 6 whereas in acetogenesis and methanogenesis step the optimum range is around 6.8 to 7.8 (Azbar, et al., 2001; Grepmeier, 2002; Vega De Lille, 2015). The pH of the pilot-plant was set once at the beginning of each batch in phase I and II to 5.5 in reactor 1 and 7.5 in reactor 2. In case of the first reactor, the pH was adjusted by adding approximately 200 ml of hydrochloric acid (15 vol%) whereas for reactor 2 about 700 ml of caustic soda solution (15 vol%) was added. The daily operation mode of such a pilot-scale treatment would require high consumption rates of auxiliary materials. For reasons of cost-effectiveness, the degradation performance was determined without having a pH-adjustment from phase III of the start-up on.

The Anammox-based third process stage (reactor 3) was inoculated with 60 liters of seeding sludge originating from a deammonification (DEMON) reactor in Fulda Gläserzell (Germany). At the
beginning (phase I) synthetic wastewater (SWW) comprising 1000 liters of tap water mixed with ammonium sulfate (40 mg/l) and sodium nitrite (50 mg/l) was fed. Afterwards a stepwise adaptation to organically degraded MWW of reactor 2 occurred in ratios of MWW to SWW of 20% to 80% (phase II), 50% to 50% (phase III) 80% to 20% (phase IV) until 100% of municipal wastewater was accomplished (phase V). Due to their mesophilic character, the optimum temperature for the Anammox bacteria is in the range of 30 °C to 40 °C (Van De Graaf, et al., 1996; Strous, et al., 1998). But further studies demonstrated also good degradation performances with temperatures around 18 °C to 20 °C (Isaka, et al., 2007; Dosta, et al., 2008). In the presented study the temperature in the Anammox-stage was kept in between 28 °C and 35 °C. The pH range of the Anammox bacteria is in between 6.7 to 8.3 (Strous, et al., 1998; Wesoly, 2009). As the previous process step of the pilot plant is in the same pH range no adjustment or even pH control is needed. After the interconnection of all three reactors, the pilot plant was tested for further 200 days of operation by feeding mechanically pretreated MWW.

Analytics
For analyzing the degradation performance of the pilot plant different online and offline measurements were carried out. First, the fluid temperature inside all reactors and the buffer tank was monitored online with Pt100 SITRANS TH400 temperature sensors from Siemens AG, Munich, Germany. As mentioned before, the tempering of the wastewater occurs only once in the buffer tank with a set point of 35 °C. Thus temperature is decreasing over time in the process units with an average of 1.125°C/h.

Additionally the pH was monitored in each of the reactors with pHD-S sc Digital Differential pH-sensors from Hach Lange GmbH, Düsseldorf, Germany. For analyzing the degradation performance of the Anammox based process a 3798-S sc Digital inductive conductivity sensor from Hach Lange GmbH (Düsseldorf, Germany) is installed in reactor 3.

In order to characterize the water quality concerning organic load, the chemical oxygen demand (COD) is measured offline via the analyzer QuickTOC® from LAR AG, Berlin, Germany. Different parameters were collected for characterizing nitrogen removal, including concentrations of ammonium-nitrogen (NH4-N), nitrite-nitrogen (NO2-N) and nitrate-nitrogen (NO3-N). The determination is done with photometric-based analytical test kits from Merck Chemicals GmbH (article-no. NH4-N: 1.00683.0001, NO2-N: 1.14776.0001 and NO3-N: 1.14773.0001).

RESULTS AND DISCUSSION

Start-up of the two-stage organic digestion
The degradation performance over 24 h retention time of the two-stage organic digestion during start-up phase is presented in Figure 3. Due to the added supplementary organic acids in the adaptation phase (phase I), the initial COD concentration in the medium was higher than usually found in domestic wastewater. Beginning with 900 mg/l in batch 1 the total COD reduction within 24 h was about 0.280 kg/(m³*d), which corresponds to a removal efficiency of 31 %. In batch 2 the removal efficiency increased to 49 % having 699 mg/l COD concentration in the inlet and a total COD reduction of 0.343 kg/(m³*d). With 598 mg/l at the beginning, batch 3 showed a total COD-reduction of 0.316 kg/(m³*d) coming up to 53 % of COD removal efficiency.

In the second phase (phase II) the SWW was replaced by the outlet of R1 to 30% (batch 4), 50 % (batch 5) and 100% (batch 6). With an inlet COD concentration of 343 mg/l and a total COD reduction rate of 0.141 kg/(m³*d) a removal efficiency of 71 % could be achieved in batch 4. In batch 5 a removal efficiency of 47 % was achieved with a starting COD concentration of 566 mg/l and total COD reduction of 0.258 kg/(m³*d). As the interconnection of the two stages was accomplished (batch 6), the degradation performance was around 0.410 kg/(m³*d) with a COD
inlet concentration of 562 mg/l, resulting in a total removal efficiency of 71 %. So, despite of changing the environmental conditions for the microorganisms, the average removal efficiency in phase II was about 63 %.

After completing the interconnection between R1 and R2, the supplemented organic acids were omitted (phase III). Although the inlet COD concentrations of 305 mg/l in average were lower than in the two phases before, the removal efficiencies for COD remained the same at around 62 %. With a total COD reduction rate of 0.198 kg/(m³*d) an average outlet COD concentration of 112 mg/l could be achieved.

Finally it is shown that the start-up of the two-stage anaerobic digestion process was successfully accomplished for a temperature range of 33.7°C and 37.5°C and without the need of a specific pH control. In the presented pilot plant COD removal efficiencies of 62 % in average were realized. Comparable results (69 % and 50 %) were achieved by Domoso-Bravo et al. (2009) in a similar lab scale plant, even though higher ammonium concentrations of about 500 mg/l were used. Although the average COD reduction rate in the pilot plant is comparatively lower than the results presented in literature (Domoso-Bravo, et al., 2009), the aimed service water limit value of 75 mg/l was almost reached even during the start-up phase.

Figure 3: Reduction of chemical oxygen demand (COD) and COD removal efficiency within 24 hours retention time during the start-up of the anaerobic digestion process. The start-up process is subdivided into following three phases: (I) adaptation of microorganisms by feeding synthetic wastewater comprising mechanically pre-treated municipal wastewater with supplementary organic acids including acetic acid, propionic acid and butyric acid in ratio of 2:1:1, (II) stepwise replacement of the synthetic wastewater by wastewater coming from reactor 1 the hydrolysis and acidogenesis stage (30 %, 50 %, 100 %), (III) operation of the interconnected two-stage anaerobic digestion without feeding additional organic acids. According to the Bavarian bathing water regulations the limit value for COD is about 75 mg/l (BayBadeGewV., 1993).

Start-up of the Anammox-based third stage

The degradation performance of the Anammox process within six hours of retention time during start-up is shown in Figure 4. Feeding 100 % of SWW (phase (I)) resulted in an average ammonium degradation rate of 0.155 kg/(m³*d) with removal efficiencies in between 90 % to 80 %. The average ammonium outlet concentration in this adaptation phase was around 7.7 mg/l. In the second phase in which 80 % of SWW and 20 % of MWW were fed, starting concentrations of ammonium were about 40 mg/l. Having removal efficiencies in between 67 % and 72 %, the average ammonium outlet concentration was about 12 mg/l. An average ammonium degradation rate of 0.113 kg/(m³*d) was reached. By increasing the part of MWW to 50 % (phase (III)) and 80 % (phase (IV)) in feed, the removal efficiencies decreased from 72 % to 47 % while initial ammonium concentrations were increased from 40 mg/l to 84 mg/l. The average outlet concentrations were 20 mg/l in phase (III) and 40 mg/l in phase (IV), resulting in ammonium
degradation rates of about 0.158 kg/(m³*d) in phase (III) and 0.150 kg/(m³*d) in phase (IV). Feeding 100% of MWW (phase (V)) the initial ammonium concentration increased further up to 82 mg/l, whereas the removal efficiency stayed between 38% and 50% and the average outlet concentrations was about 42 mg/l. The resultant average ammonium degradation rate was about 0.141 kg/(m³*d).

The reason for the decrease of ammonium removal efficiency in phases (III) to (V) is attributed to an inappropriate nitrite-nitrogen (NO2-N) and ammonia-nitrogen (NH4-N) ratio. As recommended in literature the ratio of NO2-N/ NH4-N should be in between 1.15-1.3 (Van De Graaf, et al., 1996; Van der Star, et al., 2007; Ali, et al., 2015). After increasing nitrite starting concentration up to a ratio of NO2-N/ NH4-N of 1.15-1.3, the removal efficiencies increased again to 84.1 %, 95.0 % and 97.2 % (phase VI) with an average ammonium degradation rate of 0.189 kg/(m³*d).

In order to avoid both excess substrate supply and substrate limitation during the treatment of organically pretreated municipal wastewater, the substrate to feed ratio had to be further optimized. Thus different initial NO2-N/ NH4-N ratios were tested over the hydraulic retention time. Most steady values were found for an initial ratio of 1.14 (see Figure 5, batch 16).

Finally it is shown that the start-up of the Anammox-based process was successfully accomplished.
within four weeks for the treatment of domestic wastewater in a temperature range of 28°C to 35°C and a pH range of 7.1 to 8.2. Thereby an average ammonium removal efficiency of even 92% was achieved for ammonium-poor wastewater at the end of the start-up. In literature comparable values of 85% to 99% were reported for ammonium-rich wastewater (Fux, et al., 2002). While the average ammonium reduction rate of the pilot plant was relatively low compared to the results received by Fux et al. (2002), the self-defined service water limit value of 10 mg/l could already be reached during the start-up phase.

Degradation performance of the decentralized pilot plant for the anaerobic treatment of domestic wastewater

After the start-up, the pilot plant was studied for further 200 days of operation. Figure 6 and Figure 7 show the average COD and NH4-N concentrations in the inlet municipal wastewater, after the anaerobic digestion (outlet of reactor 2) and after the Anammox-based process (outlet of reactor 3). Furthermore also the average removal efficiencies of COD and NH4-N are illustrated for the anaerobic digestion and the Anammox-based process step.

Beginning with an average COD concentration of about 212 mg/l in MWW the two stage anaerobic digestion reduces the COD concentration up to 85 mg/l resulting in an average COD removal efficiency of 60%. In reactor 3 another 21% of COD is removed to an average outlet COD of 39 mg/l. The overall COD removal efficiency of the three-stage anaerobic treatment plant during these 200 days of operation is about 81%. Considering the self-defined service water limit value of 75 mg/l for COD, solely the two-stage anaerobic digestion could not satisfy it completely. But with the downstream Anammox-based reactor, the limit value could always be hold.

In the incoming municipal wastewater the NH4-N concentrations were 46 mg/l in average. During the anaerobic digestion the ammonium concentration increased in average to 50 mg/l which can be explained by the hydrolysis of protein-rich wastewaters (Grepmeier, 2002). Only in the Anammox-stage the ammonium concentration decreased in average to 1 mg/l, resulting in. With an ammonium removal efficiency of about 96% within six hours of retention time, the self-defined service water limit value of 10 mg/l of NH4-N could even be undershot.

**Figure 6:** Average organic degradation performance of the pilot plant treating municipal wastewater (MWW) within 200 days of operation. The average COD concentrations for the incoming MWW, for the outlet of R2 after the anaerobic digestion and for the outlet of R3 after the Anammox-based process are plotted. Furthermore the COD removal efficiency for the anaerobic digestion as well as for the Anammox-based process are shown.

**Figure 7:** Average ammonium degradation performance of the pilot plant treating municipal wastewater (MWW) within 200 days of operation. The average NH4-N concentrations for the incoming MWW, for the outlet of R2 after the anaerobic digestion and for the outlet of R3 after the Anammox-based process are plotted. Furthermore the NH4-N removal efficiency for the anaerobic digestion as well as for the Anammox-based process are shown.
CONCLUSION
The presented study describes the successful start-up of a decentralized anaerobic pilot plant for production of service water out of domestic wastewater. For start-up all reactors were operated separately at first in order to facilitate the adaptation of the microorganisms to their new surrounding conditions.

The two-stage anaerobic digestion was operated in a temperature range of 33.7°C and 37.5°C and without any pH-control. Already during its start-up phase, an average COD removal efficiency of 62 % could be reached by reducing an average inlet COD concentration of 305 mg/l to 112 mg/l. Average COD removal efficiencies of 60 % in the subsequent testing phase of 200 days of operation show that the process was also successfully stabilized. Achieving an effluent COD concentration of approximately 85 mg/l, the two-stage treatment could not completely satisfy the self-defined service water limit value of 75 mg/l solely. Only by connecting the Anammox reactor in downstream, the limit value could even be undershot (39 mg/l).

Ammonium removal efficiencies in the Anammox process were in between 90 % and 79.8 % while feeding 100 % SWW. Substrate limitation led to low removal efficiencies during adjustment to municipal wastewater. Therefore the optimum NO2-N/ NH4-N ratio for the Anammox-stage of the presented pilot plant was determined to 1.14 and the nitrite feeding was adapted to this optimum ratio. Subsequently the ammonium removal in 100 % of MWW was about 95 % and could even reach the self-defined limit value for service water of 10 mg NH4-N/l with a retention time of six hours. Showing an average removal efficiency of about 96 % during the subsequent 200 days of operation, it can be reasoned that also the Anammox-process was successfully stabilized.

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