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Sand and activated carbon filtration in removing microorganisms from wastewater

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ABSTRACT

This work aimed to test the efficiency of associating two natural, local, abundant and eco-friendly granular media; sand provided from South Algeria and granular activated carbon (GAC) manufactured by Algerian Company as a tertiary treatment for urban wastewater in Algeria. The effective diameter was 0.5 and 1.2 mm for sand and GAC respectively. The pilot consisted of two circular columns; one filled with sand and the other with GAC. The raw water used was brought from the secondary treatment (activated sludge) from the Ain El Houtz wastewater treatment plant. The sand filter column was fed 5 h/d with a constant filtration rate of 3.8 m/h, then 5 L of filtered water fed the granular activated carbon filter with a filtration rate of 0.8 m/h. After 16 weeks of operation, total germs, total coliforms, fecal coliforms, *E. coli* and fecal streptococcus had been reduced in a range of 82%–90% by filtration on the sand and 92%–96% by activated carbon. *Clostridium* has been reduced by 97% and 99% by sand filtration and activated carbon respectively and *Salmonella* by 66% and 94% on the sand and activated carbon respectively. This process is promising as a tertiary treatment in terms of cost and sustainable development since only natural, local and eco-friendly materials are used.

Keywords: Sand; Activated carbon; Microorganisms; Pathogens; Filtration

1. Introduction

Water is an important natural resource used by humans; for domestic, agricultural, and industrial purposes. Water is negatively affected by diverse pollutants including physical (conductivity, total dissolved solids, and suspended solids), chemical (minerals, carbon, dissolved oxygen, nitrogen, and phosphorus), and biological (viruses, bacteria, algae, protozoan, nematodes) ones which deteriorate the quality [1]. Wastewater treatment plants (WWTP) are designed to reduce pollution by removing the organic load, solids, nitrogen, and phosphorus, however, with less attention to the microbiological issue [2].

Water demand has increased in recent years, largely due to a lack of water resources and inadequate economic structures, particularly in arid and semi-arid countries where more and more reclaimed water will be used in the future for irrigation [3]. Among the reclaimed water, there

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are non-conventional resources such as treated urban wastewater, however, before use, we must be sure that it is safe to avoid damaging public health and the environment [4].

Microorganism pollution is one of the dangerous contaminants in the water, and to remove these contaminants many disinfection techniques are applied such as; ozonation, ultraviolet, and oxidation, however, chlorination is the most widely used means to inactivate pathogenic microorganisms in water and wastewater in the world, but its effectiveness is reduced by suspended solids, turbidity, and nitrogen compounds, and also the use of chlorine in wastewater gives undesirable by-products suspected to be hazardous to humans and the environment [4].

Diverse kinds of pathogens are found in wastewater, however, impractical to monitor for assessment of a wastewater treatment system. The microorganisms that are used to assess wastewater quality are called 'indicators of fecal contamination', among these indicators; coliform bacteria, quantified either as total coliforms or fecal coliforms, fecal streptococcus, *Escherichia coli*, *Clostridium*, and *Salmonella* [1].

Sand filtration is generally considered one of the most efficient and favorable technology for the reduction of pathogens, particulate organic substances, and turbidity [5,6]. It is also an ideal treatment technology for developing countries and rural communities, where low cost, ease of operation and maintenance and removal of pollutants are of primary consideration [7].

Sand filtration could be used as a tertiary treatment option for secondary effluents on municipal wastewater treatment plants (WWTP) to reach an appropriate quality for the safe reuse of water in irrigation. However, it requires sand with specific properties regarding grain size diameter and uniformity coefficient, which might be not locally available in some regions [8].

Adsorption is often used at the end of a treatment sequence as a tertiary treatment due to a high degree of purification that can be achieved. Activated carbon is the most popular adsorbent used for the application of adsorption technique, it is considered as a heterogeneous material (made of wood, coconut shells, coal) with unique adsorptive characteristics mainly influenced by the porous structure, pore surface area (800–2,500 m^2/g) and chemical structure of the surface, giving an exceptional ability to adsorb gases, liquids and other kinds of materials on its surface [7]. It is also used to purify contaminated water and wastewater, and kill bacteria. Two types of activated carbon include; powdered or granular (used in columns filter) [9].

The present study aimed to test the efficiency of coupling two natural eco-friendly granular media; sand and activated carbon, on microorganisms removal as a tertiary treatment on urban wastewater from Ain El Houtz WWTP (Tlemcen, Algeria). The experiment was performed for 16 weeks, where water passed through a sand filter (SF) (Pilot TE400 manufactured by Deltalab, Germany) then on a granular activated carbon filter (GACF) (Polyethylene column). To monitor our study, microbiological properties of water were measured before and after both SF and GACF, those parameters were mainly bacteria indicators for fecal contamination; total coliforms (TC), total germs (TG), fecal coliforms (FC), fecal streptococcus, *Escherichia coli*, *Clostridium*, and *Salmonellas*. The assessment of the experiment was to compare our results with the quality standards of reused wastewater for irrigation in Algeria and to determine whether the removal efficiency of SF and GACF in reducing pathogens is effective and if it presents synergies when both filtrations are associated.

2. Material and method

2.1. Pilot description

The pilot that we have used for filtration on sand is the Pilot TE400 manufactured by Deltalab (Germany) (Fig. 1) and provided by the Laboratory of Valorization of Water Resources in Algeria, it is composed of a feed tank with a capacity of 150 L (1), a filtration column (2) in Altuglas (France) with an internal diameter of 100 mm and a height of 1,000 mm and stop grids with a mesh of 0.5 mm, and two brass support and stop grids with a mesh of 0.5 mm; two manual (3), 12 piezometric (4) multi tubes placed at different heights, float flow-meter (5) to control the outlet (filtered) flow and a supply pump (6) from the tank to the filter column. For the filtration on activated carbon, a second polyethylene filtration column was used with an internal diameter of 40 mm and a height of 1,000 mm, equipped with a filtrate outlet valve.

2.2. Filter media

The sand and granular activated carbon were sourced from a local Algerian supplier. The sand was brought from the Southern Algeria site and granular activated carbon (GAC) was bought from a local company SARL PROCHIMA MAGHREB (Algeria). The characteristics of the media were determined at the laboratory (Table 1).

The particle size analysis was carried out according to Standard NF EN 933-1 [10], which consists of consecutively passing a quantity of the sample through multiple sieves with decreasing diameters by applying vibrations. From the obtained grain size distribution curve we estimate the effective diameter D_{10} (where 10% of the sand particle are less than D_{10}) and D_{60} (where 60% of the sand particles are less than D_{60}), and the uniformity of the grain size distribution (the uniformity coefficient Cu) is deduced by calculating the ratio D_{60} and D_{10} [11].

The ideal sand size for a filter to have an adequate hydraulic conductivity and to minimize the risk of clogging is when D_{10} is between 0.3 to 1.5 mm (0.5 and 1.2 mm for sand and activated carbon respectively in our study) and Cu less than 4 (2.5 and 1.6 for sand and activated carbon respectively in our study) [12,13]. Two other parameters were determined at the laboratory according to Liénard et al. [14] such as the volumic mass (ρ), and the density (d).

2.3. Filtration procedure

The raw water that we have used in our study is wastewater (Table 2) from Ain El Houtz (34°55′ North, 1°19′32″ West) wastewater treatment plant (Algeria, Tlemcen), which treats 30,000 m^3/d of municipal wastewater by the activated sludge process. At present, no tertiary treatment is applied, which means that the secondary effluent cannot be used for reuse in irrigation.

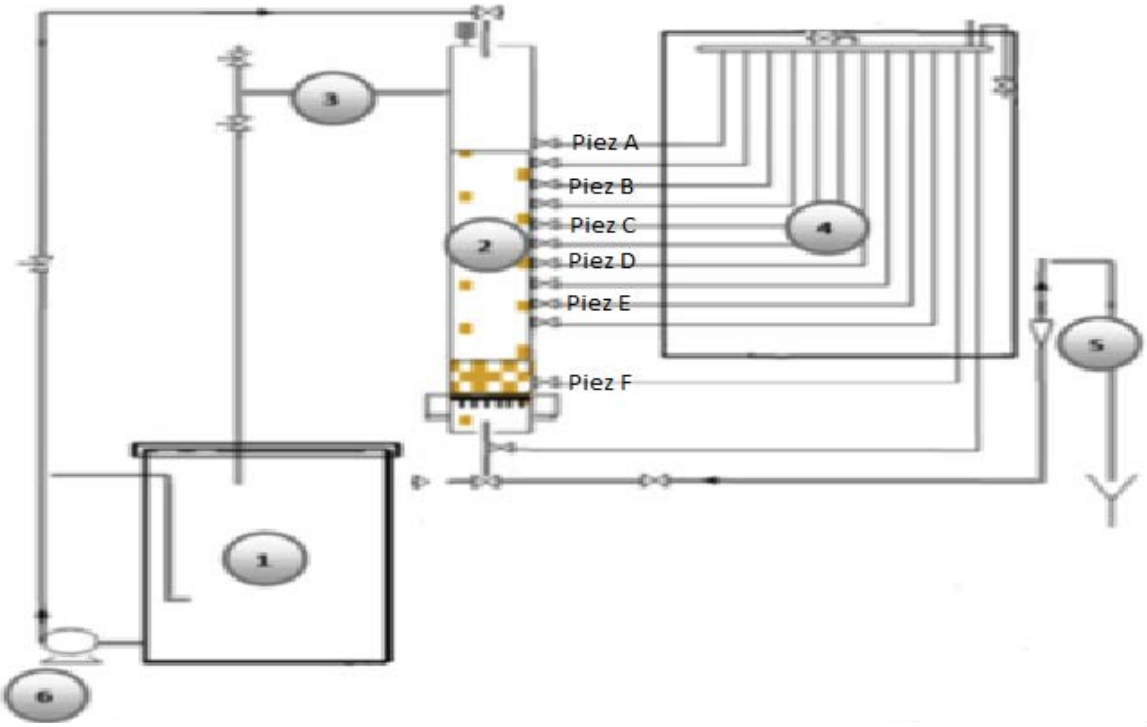

Fig. 1. Pilot TE400 (manufactured by Deltalab, Germany).

Table 1
Characteristics of the media used

	Sand	GAC
D_{10} (mm)	0.5	1.2
D_{60} (mm)	1.4	1.9
C_U	2.5	1.6
Real density	2.5	1.1
Apparent density	1.7	0.5
Conductivity (μS/cm)	3,000	700
pH	8.4	8

Table 2
Microbiological characteristics of the used raw water

Parameters	Values (CFU/100 mL)
Total germs	1,340–1,350
Total coliforms	150–164
Fecal coliforms	115–120
Escherichia coli	90–96
Fecal streptococcus	225–231
Clostridium	340–348[a]
Salmonella	15–20

[a]CFU/20 mL

The water has been taken daily from the settling tank after the secondary treatment, where temperature, pH, and turbidity were measured in situ, it varies from 20°C to 28°C, 6.4 to 8.1, and 49 to 62 NTU, respectively.

For our study, the same procedure has been followed as in Bouchenak Khelladi et al. [15] where the raw water passed through two filters; the first one was the sand filter then the second one the activated carbon filter. The filter depth is 60 and 100 cm for sand and activated carbon respectively.

To ensure optimal conditions of operation of the filtration column especially the regulation of biomass and oxygenation, the sand filter was fed for 5 h with a filtration rate of 3.8 m/h five times a week (we worked 5 d/7, and therefore a daily volume of 150 L which is the useful capacity of the feed tank) [16]. Our study took place over sixteen weeks, where at the end of each 5 h sand filtration cycle, 5 L of water is collected and then filtered by GAC. Appendix 1 summarizes the filtration procedure of our study.

2.4. Sampling and analysis

The samples were collected weekly during the 16 weeks of experiments at the inlet and outlet of each filter (SF, GACF). Samples were collected in sterile plastic sample bottles and immediately placed in a cooler box containing ice packs for processing.

The main parameters that have been used to evaluate the performance of sand and activated carbon filters are: Bacteriological parameters enumerated were TC, TG, FC, fecal streptococcus, *Escherichia coli* (*E. coli*), *Clostridium* and *Salmonellas*. All the bacteriological analysis was performed according to APHA standards (American Public Health Association) [17].

- *Total germs (TG) tested on tryptone agar medium*: It consists of the estimation of the total number of germs in water

known as the aerobic revivable bacteria that include all the aerobic bacteria, yeast, and fungi that form colonies on a specific culture medium. The medium used is tryptone agar with yeast extract composed (g/L) of tryptone 5; glucose 1; yeast extract 2.5; agar 15; completed with distilled water at 1,000 mL. The pH is adjusted at 7.0 ± 0.2 before autoclaving at 120°C for 20 min. We take two Petri dishes where we add 1 mL of water to be analyzed in each one, then the tryptone agar medium. The mixture is slowly mixed and let rest. The incubation is done for 48 h at 37°C for the first Petri dish and at 22°C for the second one.

- *Total coliforms (TC) and fecal coliforms (FC) tested on bromocresol purple lactose agar medium (BCPL)*: The bromocresol purple lactose agar medium is composed of (g/L): peptone 5; meat beef extract 3; lactose 10; bromocresol purple 25 × 10^{-3}. The pH is adjusted to 6.8 before autoclaving at 120°C for 20 min. Three dilutions have been performed 10^{-1}, 10^{-2} and 10^{-3} from the sample to inoculate the medium. In each dilution, we have inoculated three tubes by adding 1 mL of BCPL, then put them in the oven at 37°C for 24–48 h.

The first lecture has proceeded after 24 h of incubation, the tubes are considered positive if the liquid in the tube is cloudy with a variation from purple to yellow with a gas release. The number of TC per 100 mL is obtained by comparing the number of positive tubes according to the table of the most probable number (MPN). This test is called presumptive.

2.4.1. Subculturing on Schubert medium

The medium is composed (g/L) of: tryptophane 0.2; glutamic acid 0.2; magnesium sulfate 0.7; ammonium sulfate 0.4; sodium citrate 0.5; sodium chloride 2; tryptone 10; mannitol 7.5. Each positive tube (yellow variation and gas release) from the previous step is subcultured (6 drops) in the tubes with Schubert broth, then incubated at 44°C for 24 h. We consider as a positive test, the tubes where a bacterial growth appeared (cloudy) with a gas release. The FC counting is performed the same as TC and expressed per 100 mL of the sample.

- *Escherichia coli tested on BCPL and Kovacs reagent*: within the same tubes where TC and FC were tested, 2 to 3 drops of Kovacs are added in the tubes considered as positive. We consider positive tubes, the ones where a red ring appears on the surface which indicates the production of indole. The *E. coli* counting is performed referring to the MPN table in 100 mL of the sample.
- *Fecal streptococcus tested on Rothe medium*:

Presumptive test: The inoculation is done on Rothe medium composed (g/L) of bio-polytone 15.0; meat beef extract 4.5; glucose 7.5; sodium chloride 7.5; sodium acid. The pH is adjusted at 7.2 before autoclaving at 120°C for 20 min. The incubation is done at 37°C for 24–48 h. The tubes are considered positive and can be subjected to the confirmatory test if a microbial cloud appears with the settlement of the pastille on the bottom of the tubes.

Confirmatory test: We take some drops from the tubes containing the sample that we put on other tubes filled with Litsky medium composed (g/L) of peptone (Bio-Lysat) 20.0; glucose 5.0; sodium acid 0.2; purple-ethyl 0.5; NaCl 5.0; potassium hydrogen phosphate2.7; potassium dihydrogen phosphate 2.7. The pH is adjusted to 6.8 before autoclaving at 120°C for 20 min. The incubation is done at 37°C for 24–48 h. The apparition of a microbian cloud confirms the presence of fecal streptococcus. The SF counting is performed according to the MPN table.

- *Clostridium tested on meat liver agar medium*: The meat liver agar medium is composed (g/L) of liver meat 30; glucose 2; agar 6. We add distilled water to 1,000 mL. The pH is adjusted to 7.4 before autoclaving at 120°C for 20 min. Then 4 mL of the sample to be analyzed is introduced in 5 tubes that we place in a water bath at 80°C for 5 min; then cool it at 55°C. Then we add 2 drops of iron alum and 4 drops of sodium sulfite, and we fill the tubes with the meat liver agar. The incubation is done at 37°C, the first lecture is done after 24 h, and the second one after 48 h according to the MPN table.
- *Salmonella tested on the Salmonella, Shigella agar medium (SS):* The SS medium is composed (g/L) of peptone 5; meat extract 5; lactose 10; sodium citrate 10; iron citrate 3.1; bile salt 8.5; brilliant green 3.3; neutral red 25; sodium thiosulfate 8.5; agar 12. The pH is adjusted at 7.3 before autoclaving at 120°C for 20 min. The Petri dishes are incubated at 37°C for 24–48 h. The counting is performed according to the MPN table.

NB: The MPN table is in Appendix 2.

2.5. Filter biofilm

The head losses are an important indicator of filter performance which determines the operating time of a filter [18], for that reason the system was monitored daily by reading directly the head losses in the piezometers.

Head loss increases because of the filter maturation (growth of the biofilm) and particle trapping [19], and once the head loss becomes excessive and the filtration rate becomes slow, the biofilm has to be removed [20] by scraping about 2 cm from the surface of the sand bed [21].

However, in our study we did not wash the filters for two main reasons; the first one because of technical issues, it was impossible to scrap the surface of the filter since it was contained on a column, the second one because we wanted to benefit from the filter clogging which allows a better microorganisms retention and also keep the biofilm layer, which has been well established by Verma et al. [22] that the removal of the biofilm affects bacterial reductions. In this case, the filter will operate by a declining filtration rate since the clogging occurs. The filtration on sand that has been done is characterized as a fast filtration (3.8 m/h), however, as the running of the filter it will tend to turn to a slow filtration due to the clogging and the fact that the filter will not be washed.

3. Results and discussion

3.1. Evolution of head loss

The head loss increased during the experiment (Fig. 2), which lead to the reduction of the filtration rate from 3.8 to 1.9 m/h after 12 weeks of operation, the same thing has also been observed by Verma et al. [22] who reported that maintaining the filtration rate constant during filtration became so difficult. The head loss evolution according to Rolland et al. [23] is due to the solids accumulation of the filter bed during filtration, which can be very fast; in just a few weeks.

The clogging of the filter was expected in our study because we decided not to wash it, this decision was supported by the fact that we wanted to increase the microorganisms retention by letting the diameter of the pores of the filter decreased, as confirmed by Stevik et al. [24] who said that there is multiple evidence that removal of bacteria is more efficient in clogged filters compared with unclogged ones, and it has also been reported by Napotnik et al. [25] that, to have high efficiency in microorganisms removal the recommended range of D_{10} is 0.15–0.35 mm and Cu less than 2, however, in our study D_{10} is 0.5 mm and Cu is 2.5, for that reason, we let in purpose the filter get clogged to decrease those parameters. The fact that the filter still runs even after the filtration rate decreased, is explained according to Water [26] that a bigger effective size (0.45 mm in his study) minimizes the clogging, which is also confirmed in our study.

3.2. Total coliforms removal

Total coliforms are a good indicator of water contamination [7] and also the most common indicator organism for pathogens removal in WWTP, among these coliforms, we find *Escherichia coli*, *Citrobacter*, *Klebsiella*, and *Enterobacter* belonging to the family Enterobacteriaceae [1].

During the first 2 weeks of the experiment (Fig. 3), the total coliforms removal was insignificant for both SF and

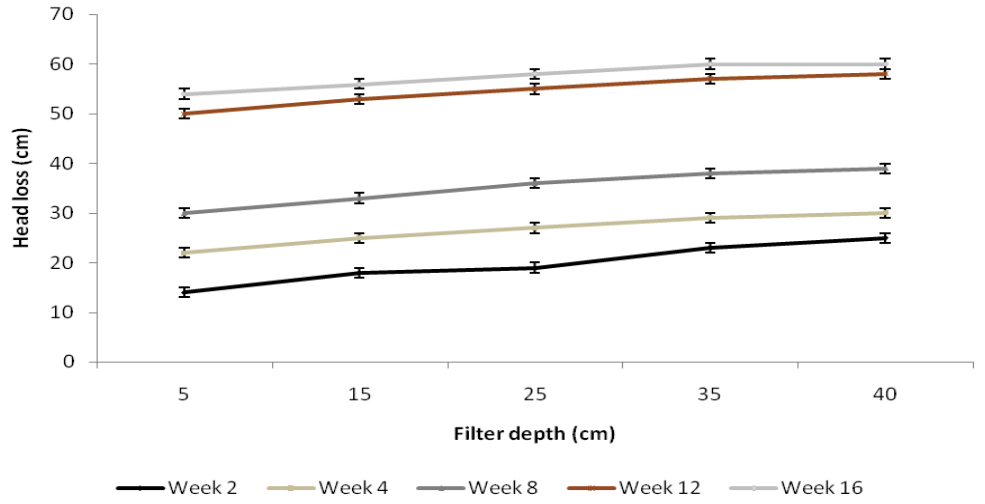

Fig. 2. Head loss evolution at different depth during 16 weeks.

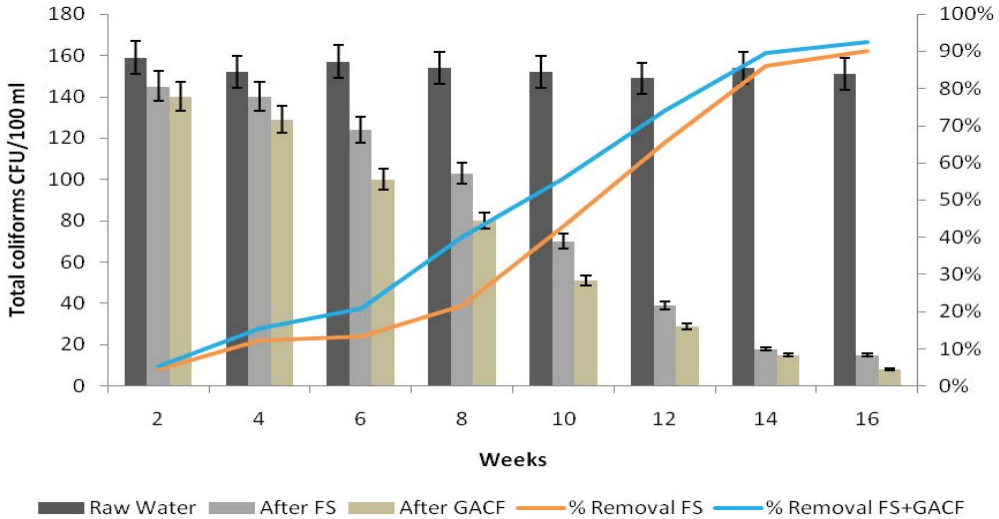

Fig. 3. Total coliforms removal during SF and GACF with time.

GACF, similar to the findings reported by Gherairi et al. [27] and Ranjan & Manjeet [21], who both attributed this period as the filter maturation also called 'the ripening of the filter', which usually takes one to two weeks, where the particle accumulates and microorganisms grow in the most top layer of the media bed (sand and GAC) as filtration progresses, during this period the filter does not effectively remove bacteria. From 3rd to 12th week, TC decreased from 163 to 39 CFU/100 mL and 143 to 29CFU/100 mL in SF and GACF respectively, which is due to the accumulation of organic particle on the filter material leading to the formation of a sticky layer consisting of bacteria and other microorganisms [28] called "*Schmutzdecke*" which provides an adsorptive surface for the attachment of organic matter and microorganisms in the water [29].

Radhi and Borghei [9] have found that by increasing the contact time, the bacteria removal increases also, and that after 90 d (14th weeks), they have noticed a significant improvement in coliforms removal efficiency up to 44%, however, in our study, it has been reached after only 60 d (9th weeks) for both SF and GACF, this difference in results could be due to the fact that we did not clean the filters.

From the 13th to 16th week, there is a slow TC decreasing in both SF and GACF, which can be explained by the 'fatigue of the filter' which decreases the performance of the filter by the appearance of the phenomenon of filter clogging [27].

The TC removal during the 16 weeks (112 d) of the experiments has reached 90% and 95% for SF and GACF, similar results have been observed by Radhi and Borghei [9], where the higher TC removal efficiency is observed at 110 d. Many studies' results agree with the results that we have found [30,31]. Finally, the percent of TC removal improves as the media bed matures, which can reach more than 99% for coliform bacteria removal [21].

3.3. Total germs removal

Total germs are defined as all the bacteria in water. During the first 6 weeks of the experiment (Fig. 4), slow TG removal was observed from 1,340 to 1,242 CFU/100 mL and 1,340 to 1,195 CFU/100 mL for SF and GACF respectively, due to the time of filters maturation. Then for the last 10 weeks, a significant improvement in TG removal has been achieved, according to Hammes et al. [32], there are two mechanisms for germs retention and inactivation; a physical mechanism which is involved in straining and adsorption of microorganisms which occurs once the filter has retained particles on its surface leading to pore size reduction, and biological mechanisms which is the interaction of pathogens with biofilm formed on the sand particle, where predation is responsible for removal and inactivation of microorganisms.

TG removal during the 16 weeks of the study has reached 89% and 93% for SF and GACF respectively. The activated carbon allows a good site for the bacteria to adsorb on its surface [33], which can be influenced by the organic matter, biofilm composition, and electrostatic attraction [24,34].

3.4. Fecal coliforms removal

Fecal coliforms are the most commonly used indicators to evaluate the level of fecal contamination and the efficiency of pathogen removal in sewage treatment processes [35].

An insignificant decrease of FC has been observed after two weeks of operation (Fig. 5) as seen before for TC (Fig. 3), then from the 3rd to the 16th week, we have noticed a significant FC removal with 85% and 94% for SF and GACF respectively, almost similar results have been reported by Letshwenyo and Lebogang [36], where 100% efficiency of FC removals were achieved in the 17th week of operation, attributed to the maturity of the filter beds.

3.5. Escherichia coli removal

The presence of *E. coli* in water is very significant as it indicates fecal contamination because several coliforms are present in unpolluted water and soil. *E. coli* constitutes more than 90% of the coliform flora of the human gut which will

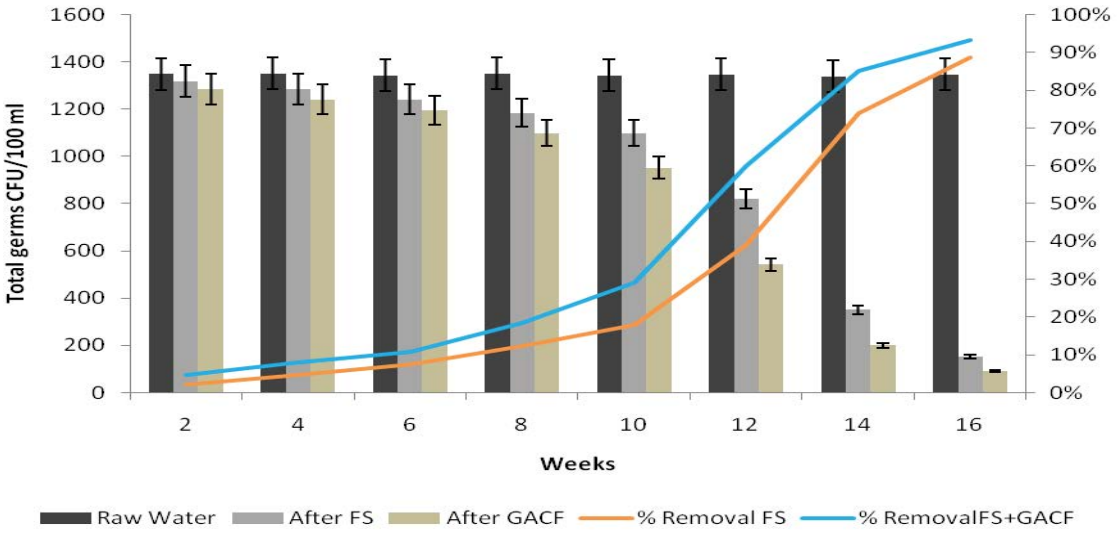

Fig. 4. Total germs removal during SF and GACF with time.

be excreted with feces and thus increases the likelihood of the presence of harmful pathogens [1,37].

After 16 weeks of operation, the *E. coli* removal rate achieved 84% and 96% for SF and GACF respectively (Fig. 6), according to Mälzer [38] and Kaetzl et al. [8], the overall *E. coli* removal is due to the contribution of the biofilm which is the main removal mechanism, confirmed also by Langenbach et al. [39], who considered that *E. coli* adsorbs much better to the biofilm which is composed of 90% organic material than to the inorganic sand grain surface and that the accumulated material improves straining and adsorption in the slimy biofilm matrix of this layer. For that reason, GACF; which is an organic material, gave better rate removal of *E. coli* than SF.

3.6. Fecal streptococcus removal

During the first 5 weeks of operation, a slight fluctuating in streptococcus removal was observed from 228–200 CFU/100 mL and 200–185 CFU/100 mL for SF and GACF respectively (Fig. 7), a similar trend has been also observed by Letshwenyo and Lebogang [36].

The average removal efficiency of streptococcus during the 16 weeks of the experiment was about 82% for SF and 92% for GACF, where other studies [36,40], showed a removal efficiency of streptococcus about 96%, higher than what we have found, this difference can be explained by the difference in the raw water quality, filter design and hydraulic loading rate used in the studies [39].

3.7. Salmonella removal

Salmonella is one of the major pathogens causing foodborne illness in developed countries. The presence of *Salmonella* in water generates a risk to public health since it represents the most frequent pathogen found in surface waters, which can be considered an important source of transmission on food via irrigation [35].

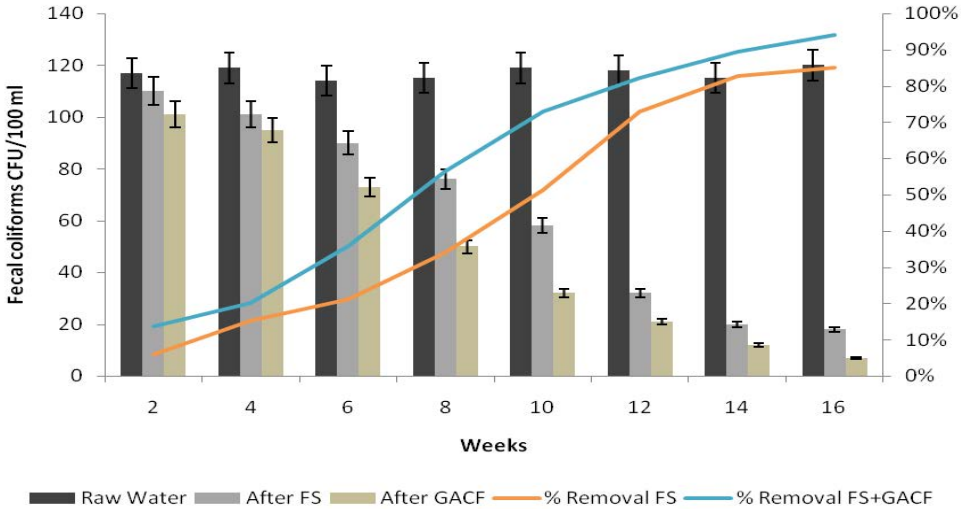

Fig. 5. Fecal coliforms removal during SF and GACF with time.

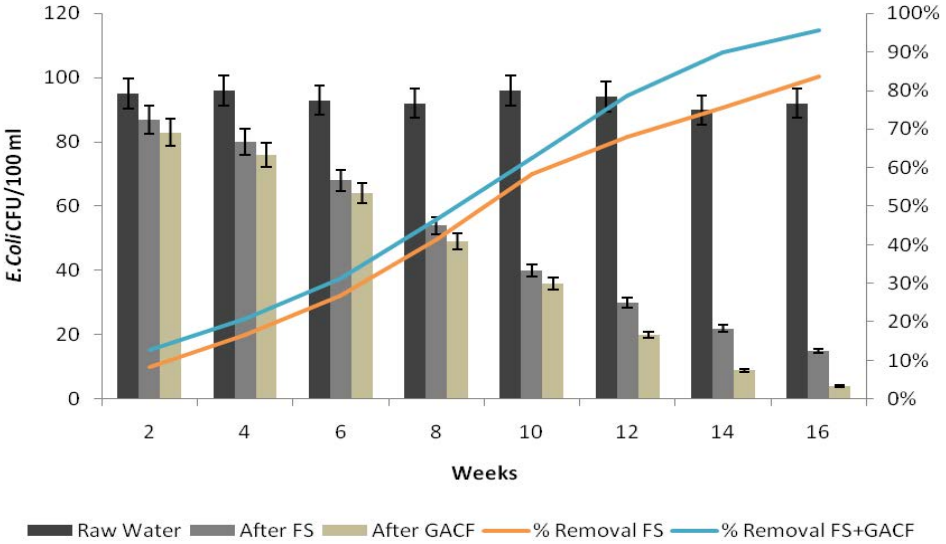

Fig. 6. *Escherichia coli* removal during SF and GACF with time.

During the experiment, 66% of *Salmonella* has been retained on SF, and 94% on GACF (Fig. 8). Activated carbon filter gave a better performance in removing *Salmonella* compared to a sand filter, which could be attributed to the large surface area (specific surface) of activated carbon supporting high biomass density (biofilm) and also precipitation/fixation reactions. Slow sand filtration for water and wastewater treatment – a review [22].

3.8. Clostridium removal

The slow decrease of *Clostridium* has been observed during the first 2 weeks of operation (Fig. 9) from 340 CFU/20 mL to 329 CFU/20 mL for SF and 340 CFU/20 mL to 325 CFU/20 mL for GACF, the same trend has been observed previously on TC (Fig. 3) and FC (Fig. 5), which has been attributed to the time of filter maturation.

At the end of the experiment of 16 weeks, *Clostridium* removal has reached 97% and 99% for SF and GACF respectively. When we compared the efficiency of SF and GACF in microorganisms removal, we have noticed that *Clostridium* removal has the highest rate, same results have been observed by Medeiros et al. [2], who attributed this trend to the fact that *Clostridium* was large enough to be retained in the filter bed.

Zhang & Farahbakhsh [41] and Kistemann et al. [42], have found that tertiary WWTP with sand filtration was able to significantly improve the efficiency of removal of

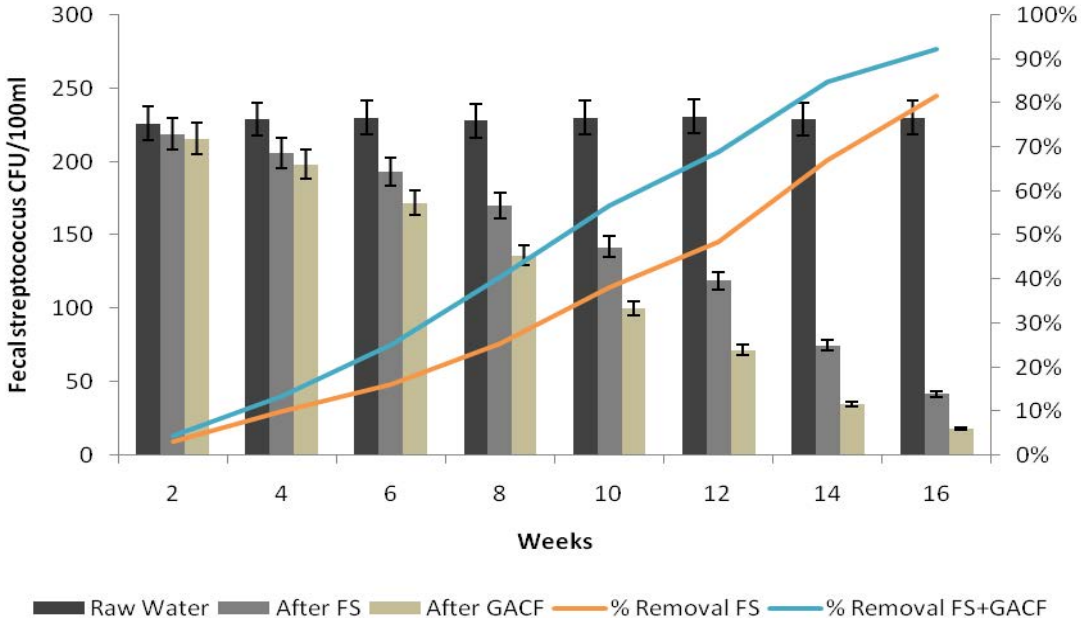

Fig. 7. Fecal streptococcus removal during SF and GACF with time.

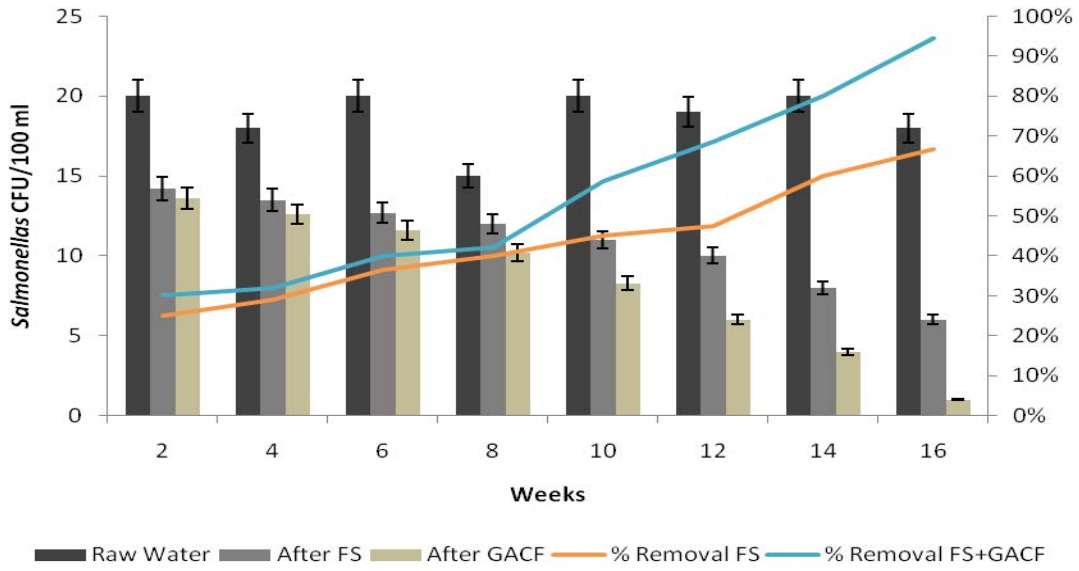

Fig. 8. *Salmonella* removal during SF and GACF with time.

microorganisms, which confirms our findings, where SF allowed a removal range of 66%–97%. The efficiency of removal of microorganisms by sand filtration is mainly due to; low filtration rate, effective sand size, and biological activity of the '*Schmutzdecke*' [2]. However, the novelty of our study was to associate sand with GAC filtration on wastewater, this association has lead to a better reduction of bacteriological parameters (Table 3) compared to SF used alone, with a range of 92%–99%, the interest of coupling sand and activated carbon is that it has enhanced bacteria removal with an average of 10%.

If we compare the wastewater quality after SF and GACF with irrigation limitation by WHO [43] (Table 4), all the parameters are under the limitations expect *Clostridium* and *Salmonella* that have to be absent in the water for irrigation which in our study it is 4 CFU/20 mL and 1 CFU/100 mL respectively. This process is very interesting and efficient as a tertiary treatment for urban wastewater for fecal bacteria removal; only local, natural, and eco-friendly materials are used, which makes it simple, accessible, and low cost.

4. Conclusion

The experiment conducted during this study aimed to test the efficiency of coupling two natural eco-friendly granular media; sand (filtration rate of 3.8 m/h) and activated carbon (filtration rate of 0.8 m/h), on pathogenic microorganisms removal as a tertiary treatment on urban wastewater from Ain El Houtz WWTP (Tlemcen, Algeria). The results obtained in the present study are very interesting because a significant improvement in pathogenic bacteria removal has been achieved.

The total germs, total coliforms, fecal coliforms, *E. coli* and fecal streptococcus had been reduced between 82%

Table 4
Limitations for wastewater reuse in irrigation [43]

Microbiological parameters (CFU/100 mL)	Limitations
Total germs	<200
Total coliforms	<120
Fecal coliforms	<100
E. coli	<100
Fecal streptococcus	<20
Clostridium	0[a]
Salmonella	0[b]

[a]CFU/20 mL; [b]CFU/5 L

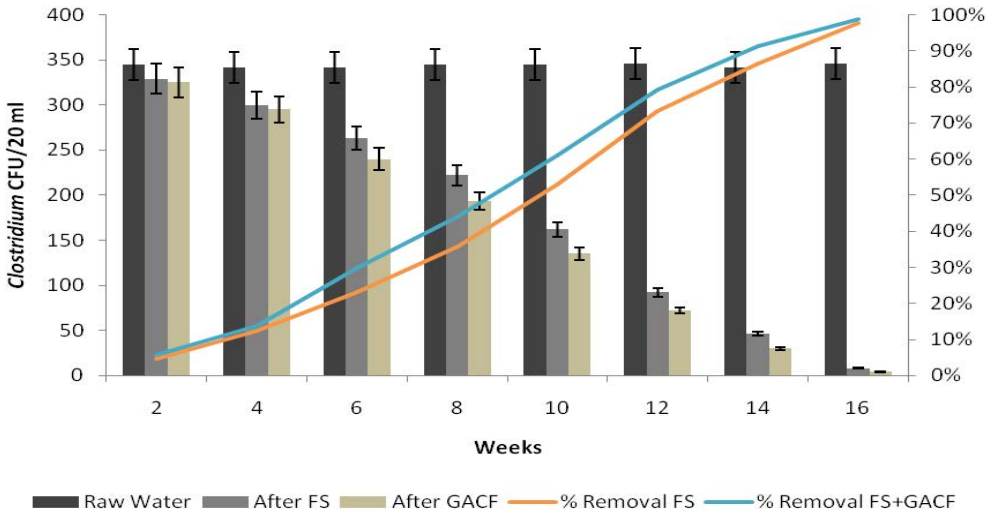

Fig. 9. *Clostridium* removal during SF and GACF with time.

Table 3
Efficiency of SF and SF + GACF in microorganisms removal

Microorganisms (CFU/100 mL)	Raw water	After SF	After GACF	% Removal SF	% Removal SF + GACF
Total germs	1,340–1,350	152	90	89%	93%
Total coliforms	150–164	15	8	90%	95%
Fecal coliforms	115–120	18	7	85%	94%
E. coli	90–96	15	4	84%	96%
Fecal streptococcus	225–231	42	18	82%	92%
Clostridium	340–348[a]	8	4	97%	99%
Salmonella	15–20	6	1	66%	94%

[a]CFU/20 mL

and 90% by filtration on the sand and between 92% and 96% by activated carbon.

Clostridium has been reduced by 97% and 99% by sand filtration and activated carbon respectively, however, it still does not conform to the norms of irrigation exceeding by 4 CFU/20 mL. Concerning *Salmonella*, it has been removed by 66% and 94% on the sand and activated carbon respectively, it is also not conforming to the WHO regulation, exceeding by 1 CFU/100 mL.

It can be concluded that by including activated carbon filtration to sand filtration, a significant improvement of more than 10% of pathogens removal has been noticed, and even if *Clostridium* and *Salmonella* exceeded the irrigation limitations, SF and GACF associated are efficient as tertiary treatment for removing bacteria when reusing wastewater in irrigation is targeted.

This process is very promising and can be easily adapted for treating urban wastewater in Algeria when reuse is intended, it is at the same time easy, low cost and fit the sustainable development; because only local, abundant, and natural materials are used not harmful neither for humans nor for ecosystems. By reusing wastewater, conventional resources are economized for domestic purposes since agriculture is the major water consumer in Algeria with about 60%.

For recommendation, chlorination could be added after the tertiary treatment to maintain a good quality of the treated water for irrigation reuse, especially when it is stored or supplied (passing through pipes) for irrigation, but also to ensure *Clostridium* and *Salmonella* removal since they were not completely removed after filtration on sand and GAC.

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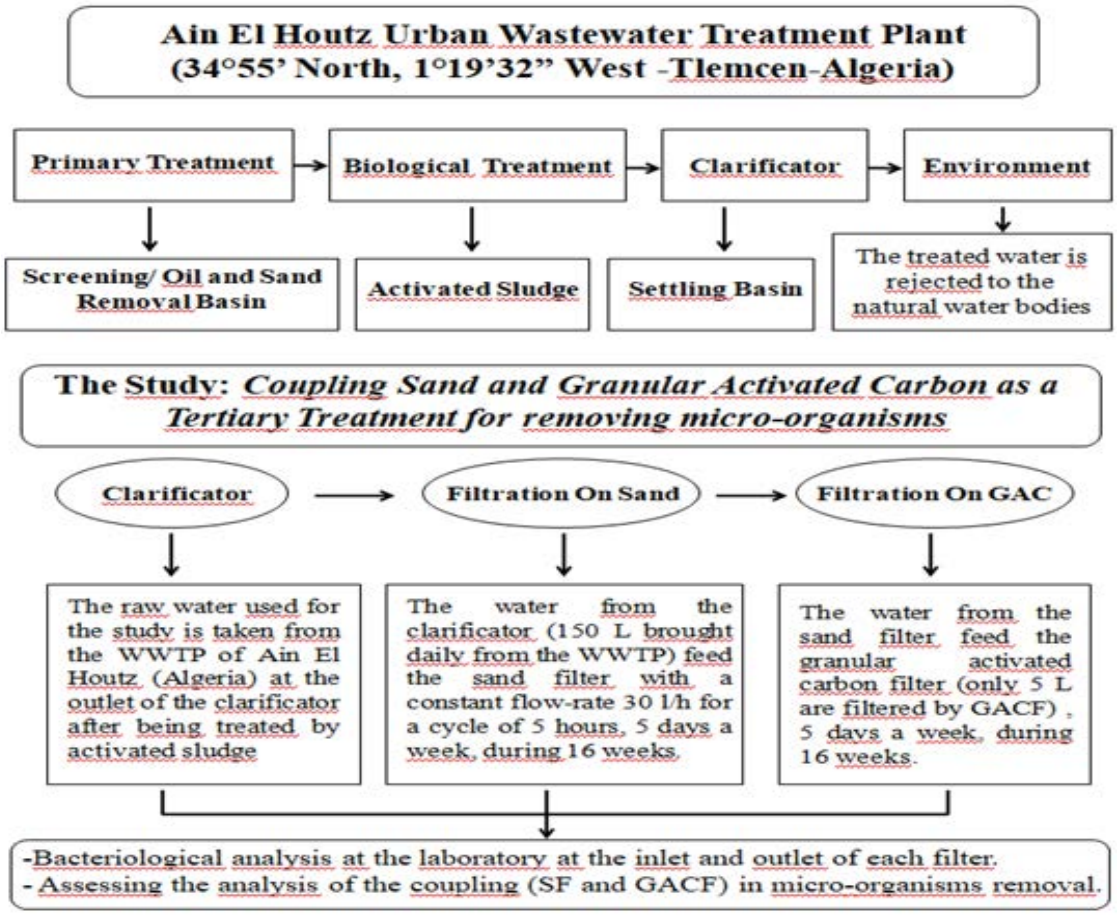
Appendices

Appendix 1: Diagram of the steps of the study



Appendix 2: MPN table used for counting (APHA Standard) [17]

MPN index and 95% confidence limits for various combinations of positive results when five tubes are used per dilution (10 mL, 1.0 mL, 0.1 mL)							
Combination of positives	MPN index/100 mL	95% confidence limits		Combination of positives	MPN index/100 mL	95% confidence limits	
		Lower	Upper			Lower	Upper
				4-2-0	22	9.0	56
0-0-0	<2	–	–	4-2-1	26	12	65
0-0-1	2	1.0	10	4-3-0	27	12	67
0-1-0	2	1.0	10	4-3-1	33	15	77
0-2-0	4	1.0	13	4-4-0	34	16	80
				5-0-0	23	9.0	86
1-0-0	2	1.0	11	5-0-1	30	10	110
1-0-1	4	1.0	15	5-0-2	40	20	140
1-1-0	4	1.0	15	5-1-0	30	10	120
1-1-1	6	2.0	18	5-1-1	50	20	150
1-2-0	6	2.0	18	5-1-2	60	30	180
2-0-0	4	1.0	17	5-2-0	50	20	170
2-0-1	7	2.0	20	5-2-1	70	30	210
2-1-0	7	2.0	21	5-2-2	90	40	250
2-1-1	9	3.0	24	5-3-0	80	30	250
2-2-0	9	3.0	25	5-3-1	110	40	300
2-3-0	12	5.0	29	5-3-2	140	60	360
3-0-0	8	3.0	24	5-3-3	170	80	410
3-0-1	11	4.0	29	5-4-0	130	50	390
3-1-0	11	4.0	29	5-4-1	170	70	480
3-1-1	14	6.0	35	5-4-2	220	100	580
3-2-0	14	6.0	35	5-4-3	280	120	690
3-2-1	17	7.0	40	5-4-4	350	160	820
				5-5-0	240	100	940
4-0-0	13	5.0	38	5-5-1	300	100	1,300
4-0-1	17	7.0	45	5-5-2	500	200	2,000
4-1-0	17	7.0	46	5-5-3	900	300	2,900
4-1-1	21	9.0	55	5-5-4	1,600	600	5,300
4-1-2	26	12	63	5-5-5	≥1,600	–	–