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SEM STUDIES ON FACILITATED MICROBIAL GROWTH ON PAC IN A BIOLOGICAL REACTOR

SALEH AL-MUZAINI
Environmental Sciences Department, Kuwait Institute for Scientific Research, P. O. Box 24885, Safat, Kuwait.

دراسات الميكروسكوب الإلكتروني في معرفة نمو البكتريا على جزيئات الكربون النشط في المفاعل البيولوجي

صالح محمد المزيني
قسم العلوم البيئية معهد الكويت للأبحاث العلمية
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لقد استهدفت هذه الدراسة استخدام جزيئات الكربون النشط مع الحمأة النشطة في معالجة المياه المستخدمة حيث أن وجود الكربون يساعد على تكاثر ونمو البكتريا عليه، لذا تم الإستفادة منه في معالجة المياه الصناعية السائلة.

وخلال فترة الدراسة تم تشغيل الوحدة التجريبية المختبرية وتزويدها بالمياه الصناعية المستعملة بمعدل يتراوح مابين ٤ – ٦ مليجرام لكل لتر تحت درجة حرارة حوالي ٢٤ درجة مئوية، أما الأوس الهيدروجيني يعادل حوالي ٧ وحدات، أما كمية الأكسجين الذائب مابين ٥ – ٦ مليجرام لكل لتر .

كما تم تزويد المفاعل البيولوجي ببكتريا من محطة تنقية المجاري الصحية لتهيئة البيئة التي تناسب الكائنات الحية الدقيقة للقيام بنشاطاتها لأكسدة المواد العضوية وتنقية المياه. وخلال تشغيل الوحدات التجريبية تم إضافة ١٢٠ مليجرام لكل لتر من الكربون النشط وتم الإحتفاظ بكمية التركيز خلال فترة الدراسة.

وأثناء التجارب المختبرية تم أخذ عينات من الكربون النشط على فترات زمنية متتالية ومن ثم تم فحصها تحت الميكروسكوب الإلكتروني للتعرف على نوعية الكائنات الحية الدقيقة، وأثبتت التجارب المختبرية من الصور الفوتوغرافية أنه هناك تنوع بيولوجي على سطوح جزيئات الكربون النشط طبقاً لفترة الحضانة، والذي ساعد بدوره في التخلص من الشوائب من المياه الصناعية السائلة.

Key words: Bioforms, Dissolved oxygen, Wastewater treatment.

ABSTRACT

A process known as powdered activated carbon (PAC) treatment was applied to facilitate additional bacterial growth in the treatment of industrial wastewater. Refinery wastewater was fed into a pilot plant reactor at a rate of 4-6 mg/l at 24°C, pH 7.0 and dissolved oxygen in the range of 5-6 mg/l. The reactor had input from a local municipal wastewater treatment plant, with the addition of 120 mg/l of PAC to the system. Carbon particles were removed from the reactor at different intervals, fixed, and examined by the scanning electron microscopy. Micrographs illustrated a variety of bioforms on activated carbon surfaces. These multiplied with incubation period, providing evidence of improved mineralization of waste.

INTRODUCTION

Shuaiba Industrial Area (SIA) which is located about 50 km south of Kuwait City accommodates 18 industries, three major refineries, two petrochemical plants, two desalination plants, an industrial gas corporation, a cement company, a paper product company, a sulphur industrial company and several others.

The SIA generates an average flow of 23,000 m^3/d and at peak about 50,000 m^3/d (1). There is little dispersion of the pollutants from the coastal area of SIA due to the shallow water conditions and the tendency of pollutants to stay and concentrate around the discharge outfalls (2). The main pollutants discharged to the SIA sea area by the existing industries include toxic organic chemicals, heavy metals such as lead, copper, nickel, vanadium and chromium, oil/grease, urea, sulphate, and phosphate (3).

The Shuaiba Area Authority (SAA) has taken steps to establish a central treatment plant to treat the combined wastewaters (industrial and sanitary) from the SIA. In order to investigate the ability of the biological treatment method (combined with PAC) to remove organic pollutants from the SIA wastewater, a pilot-scale biological reactor was constructed (4).

Benefits of a PAC-supplemented system have been reported for handling industrial discharges containing organic priority pollutants, landfill leachates, contaminated ground water, synthetic fuel production wastewater, combined municipal, industrial wastes and domestic sewage (5, 6, 7).

In this study, refinery wastewater was fed to the PAC-supplemented reactor and the growth of the microorganisms on the surface of carbon particles during the incubation period was followed. Scanning electron microscopy was used to describe the physical relationship between the carbon surface and bacterial populations, as well as the type and structural characteristics of organisms that attach to the carbon particles.

MATERIALS AND METHODS

Experimental procedure

The refinery wastewater under investigation was continuously fed into a mixed reactor with a volume of 6 liters. The reactor vessel was kept at 24°C, pH 7.0 and dissolved oxygen in the range of 4-6 mg/l with intermittent magnetic stirring. The sludge age was controlled by periodic wasting of sludge from the bottom of the clarifier. The hydraulic retention time was kept at 4 hours. Throughout the experiment, the pilot plant reactor was kept running in this manner. Fig. 1 shows a diagram of the experimental system.

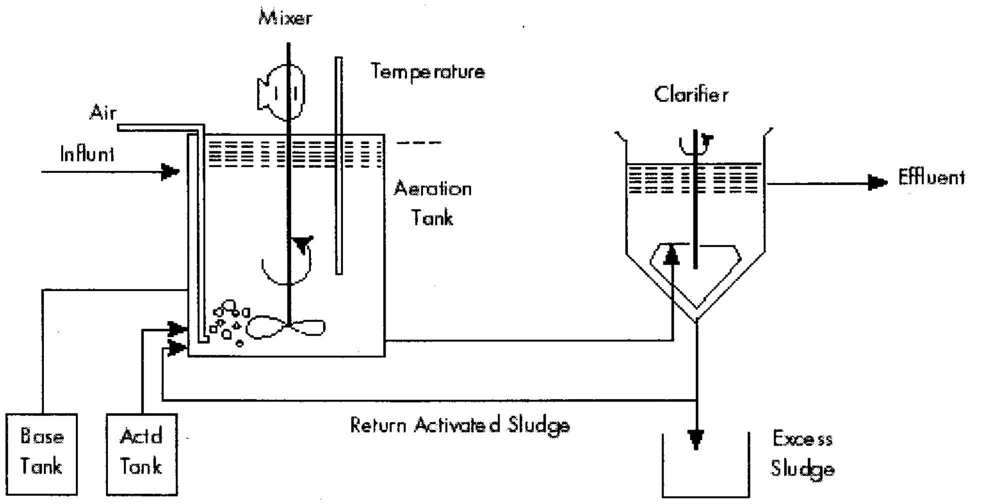

Fig. 1: Diagram of the experimental system.

Start-up of the experiment was accompanied by seeding the reactor with activated sludge (AS) collected from a local municipal wastewater treatment plant treating primarily domestic waste. The activated biomass was transferred to the pilot plant reactor and was acclimatized to the refinery wastewater under investigation for a period of 4 weeks. After acclimation, the reactor received a steady flow of 11.3 l/hr. Under the conditions 120 mg/l PAC was added to the reactor. With steady circulation time, the PAC was homogeneously distributed.

A biofilm was formed on the PAC particles. A biomass sample was taken, the carbon particles were removed, fixed and examined by scanning electron microscope (SEM) [8, 9, 10].

Samples of both virgin and biofilm-coated activated carbon were immersed for 2 hrs in 4% glutaraldehyde buffered with 0.1 m sodium phosphate at pH 7.4. Following a brief rinse in buffer at pH 7.3 [9, 6] samples were dehydrated in an ethanol series (25, 50, 75, 90%) for 10 min changes in 100% ethanol. Samples were prepared for critical point drying using liquid carbon dioxide as the transitional fluid, mounted on aluminum studs and coated with gold to a thickness of 20 mm to minimize charging and increase the conductivity of the biological materials. Samples were purged with argon gas and examined by SEM (AJEOL-EPA Model, JEOL Ltd., Japan). The SEM was operated at an accelerating voltage of 25 kv with a resolution of 10A° [11]. In order to examine the specimens for control samples (Virgin Carbon) the fixation, dehydration and drying steps were eliminated, the carbon particle samples were glued to the studs and then coated with gold (about 2 nm thick).

RESULTS AND DISCUSSION

SEM photomicrograph of the virgin carbon (Fig. 2A) was taken to provide a control which shows carbon surfaces free from bacterial growth. Fig. 2A also shows the holes and curve of site suitable for bacteria growth. The rough surface of carbon serves as an excellent shield and shelter from fluid shear forces and provides a favorable environment for bacterial growth.

Fig. 2A: Surface of virgin PAC sample without biological growth

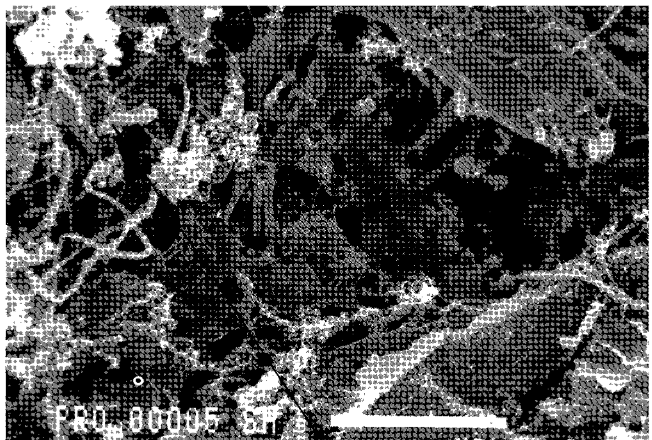

Fig. 2B: PAC particle exposed to refinery industry wastewater for 44 days (rod-shape bacteria were attached, PAC dosage 120mg/L, sludge age 3 days).

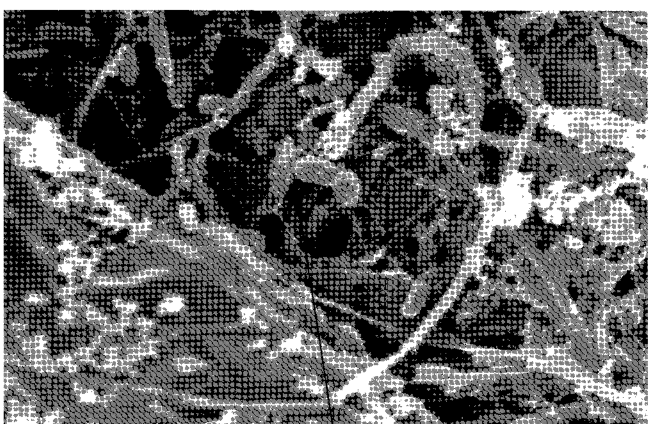

Fig. 2C: PAC particles exposed to refinery industry wastewater for 44 days (rod-shaped bacteria are present in the wastewater PAC dosage 120 mg/L; sludge age 3 days).

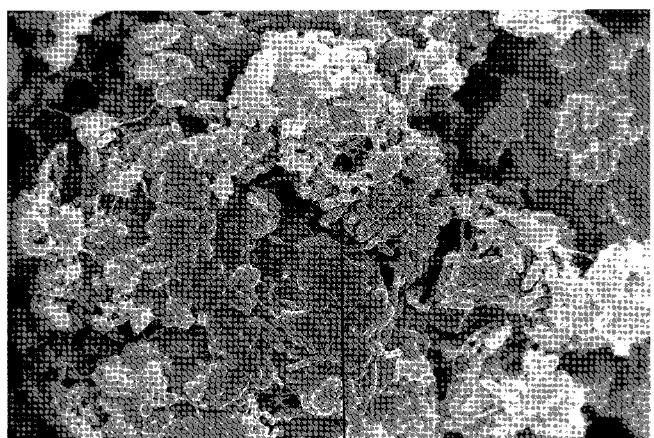

Fig. 2D: PAC particles exposed to refinery industry wastewater for 56 days in complete mixed reactor, with 120 mg/1L of carbon, (rod-shaped bacteria are growing on PAC particles. PAC dosage 120 mg/L sludge age 12 days).

Fig. 2: SEM photomicrographs of (A) virgin PAC/AS sample, 3000X; (B) 44 day PAC/AS sample, 3000X; (C) 44 day (PAC/AS sample, 5000X; (D) 56 day PAC/AS sample, 3000X; (E) and (F) 56 day PAC/AS sample, 25000X.

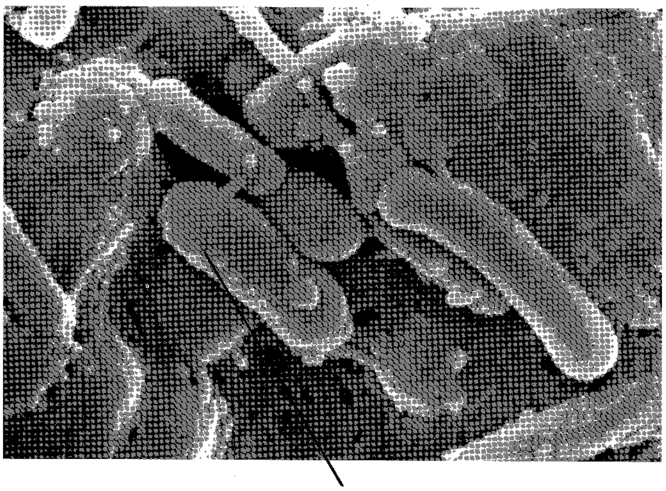

Fig. 2E: PAC particles exposed to refinery industry wastewater for 56 days in complete mixed reactor, with 120 mg/1L of carbon, (rod-shaped bacteria are growing on PAC particles).

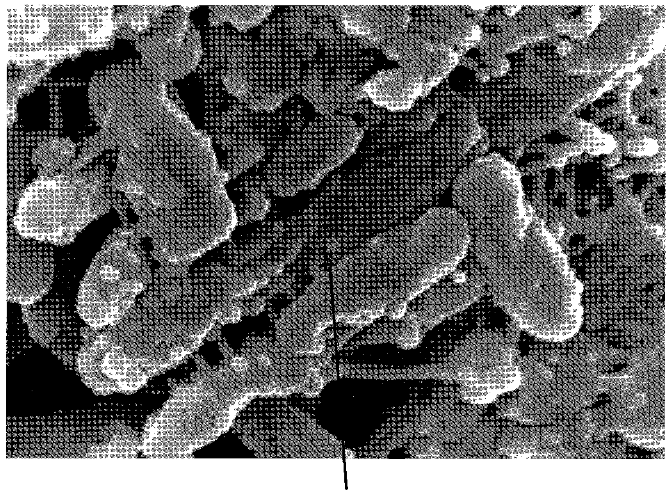

Fig. 2F: PAC particles exposed to refinery industry wastewater for 56 days in complete mixed reactor, with 120 mg/1L of carbon, (rod-shaped bacteria are growing on PAC particles).

The first SEM observation of a sample of the PAC that was in contact with the AS was taken after 44 days of operation. At the time the PAC/AS sample was collected, the PAC concentration in the AS reactor was equal to 120 mg/l and O_c (the mean cell residence time) was equal to 3 days. Two photomicrographs were taken of the same PAC/AS sample using magnifications of 3,000 and 5,000X. At a magnification of 3,000X (Fig. 2B) a variety of microorganisms are shown to be attached to the surfaces of the PAC. Also visible in this photomicrograph are PAC surfaces which are free of microorganisms. At the higher magnification of 5,000X (Fig. 2C) several rod shaped bacterial cells are shown which are attached to the PAC surfaces.

After a period of 56 days of operation another sample was withdrawn from the AS reactor and examined under the SEM. the operating conditions within the AS reactor when this sample was withdrawn were the same as the preceding sample except for a O_c equal to 12 days. An overall photomicrograph of the PAC/AS at a magnification of 3,000X is reproduced in Fig. 2D. The PAC within the photomicrograph is completely covered with a layer of rod-shaped bacteria. When the magnification was increased to 25,000X (Fig. 2E and 2F) the attachment of the individual rod-shaped bacteria to the PAC surface could be seen. It appears from this photomicrograph that not all of the PAC surface is covered with bacteria, which means that some of the PAC surfaces were available for absorption. The figures also indicated that more microbial colonization of the activated carbon surface is occurring. Thus, the present study supports earlier findings that the surfaces of activated carbon are excellent for colonization by microorganisms. Its absorptive properties serve to enrich substrate and oxygen concentrations, the rough surface provides regions that are sheltered from fluid shear forces and the variety of functional groups on the surface can enhance attachment of microorganisms [12]. This establishment of bacterial growth is evidence of improved mineralization of organics as well as nutrient regeneration [13]. In other words, the carbon surfaces enhanced the biodegradation of substrates in the media [6]. This process could be applied to a variety of wastes, including industrial discharges containing biodegradable organic substances.

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