Original Article

Investigating Effects of Clay and Carbonaceous Nanoparticles on Asphalt Degradation Aerobic Microorganisms

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Abstract

Background: Hot mix asphalt (HMA) is one of the most important structures to implement the road and street surfaces, which always exposed to destruction. Provide new solutions to reduce the destructive process of effective factors on asphalt destruction and increasing prolong of its lifetime is important for society of road engineers. Traffic loading and weather conditions are the major factors that have role on different asphalt destruction.

Materials and Methods: In this study, three types of HMAs, which contain nanoclay, and carbon nanotubes were produced by Marshall method in Iran University of Science and Technology, and one type of aged HMA were also collected for further investigations. The HMA samples were transferred to microbiology laboratory of Shahid Beheshti University of Medical Sciences, and then suspensions were prepared from specimens and cultured on nutrient agar medium. After 24 hours' incubation at 37℃ the bacteria that were grown on the plate were identified. At least the effect of clay and carbon nanoparticles on minimum inhibitory concentration (MIC) of bacteria, were determined by microdilution broth method.

Results: A lot of bacterial colonies (*Bacillus* and *Pseudomonas*) were collected from the aged specimens. The asphalt, which contain nanoclay had more bacteria than the nanocarbon asphalt. The carbon and clay nanoparticles in 2% and 4% concentration prevented the growth of bacteria, respectively.

Conclusion: The old asphalt had more bacteria than other samples, which shows bacteria are one of the main factors in decomposition of asphalt by applying corrosion to bituminous hydrocarbons. Presence of clay and carbon nanoparticles in asphalt structures could increase the durability of the HMAs and reduce the relevant economics costs.

Keywords: Asphalt, Nanoclay, Nanocarbon, Microorganisms

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Introduction

Bitumen is one of the main components of asphalt surfaces, which keeps aggregates such as sand and mineral powders beside each other[1]. In addition, bitumen can be a mixture of polycyclic aliphatic and aromatic hydrocarbons[2]. A qualified asphalt can tolerate pressure of traffic, atmospheric and environmental factors such as freeze, temperature changes[3]. According to the asphalt application, it is

divided into three categories of hot, cold and protective asphalt[4]. Cracks, holes, inconsistencies and other types of failures are results of small breakdowns. Asphalt may normally hang on for 12 to 15 years without maintenance.

In 2009, ultra violet, oxidation, heavy vehicles, water as well as oil of cars as a failure factor of asphalt were investigated. Ultra violet as an effective factor on asphalt grinding and other factors like oxidation as an important agent on the loss of gloss and glaze of asphalt, camion and heavy vehicles, water flow into asphalt structure and grease of cars can destroy the asphalt over time[5].

There are some association factors which are effective in increasing the asphalt degradation which identification and elimination of them will lead to improvement of durability of asphalt pavement, reduce costs of repair, maintenance, replacement and the possibility of further and more efficient use of asphalt pavements. One of these factors is microorganism's interactions and the road pavement structures, which can cause breakdowns such as rutting, cracking, raveling, stripping and weathering and disintegration.

Aerobic bacteria are extensively found in all environments and act as main contributing factors in decomposition of asphalt through weathering and cracking[6]. The most common microorganisms, which decompose asphalt, are from bacilli form bacteria that consist of gram-negative aerobic bacilli form such as *Pseudomonas aeruginosa* and *Bacillus subtilis* as gram-positive aerobic bacteria. *Bacillus* is a dominant microorganism of soil[7]. *Pseudomonas* is found in soil, water and other humid environments[8]. The old asphalt gradually decomposed by bacteria which produced biofilm (accumulation of bacteria which increase their resistance to antimicrobial agents). Substrate is a substance that bacterial enzymes effect on it.

In this study enzymes of microorganisms using a substrate in the bitumen structure or asphalt and able to proliferation and survivance in the asphalt structure and from this way, the physical and chemical interactions that lead to destruction structure of asphalt[9]. A nanoclay is composed of mineral silicates[10]. Carbon atoms in the walls of carbon nanotubes produce arranged and connected hexagons[11]. It has demonstrated use of nanomaterials as an additive, effects on physical properties and bitumen thermal sensitivity. Therefore, it improves the performance characteristics of the asphalt mixture[12]. The aim of this case-control study is finding a novel view to the mechanism of degradation of bitumen components and asphalt.

Methods

Manufacturing and collection of asphalt samples by using the facilities of bitumen and asphalt laboratory of University of Science and technology of Iran was performed. The models included four cylindrical 1200g samples. Among these samples two of them were simple asphalt, one other containing nanocarbon and the last sample had nanoclay.

The nanocarbon used is single walled carbon nanotubes with American COOH and the nanoclay used is bentonite nanoclay, which purchased from Arzan Azma online store. Before mixing the aggregates were heated in the oven for 24 hours at 170 ℃. Aggregates and bitumen after preparation and heating with specified values in the presence of nanomaterials in standard formats mixed with each other and by Marshall compaction hammer system, were made and dense in a standard manner. After cooling, they entered in microbial experiments. Investigation and identification of common aerobic microorganisms was performed by gram staining, as a high sensitivity, method that samples transferred from the bitumen laboratory of the Iran University of Science and Technology to microbiology laboratory of Shahid Beheshti University of Medical Sciences. Then the samples were cultured on Mueller Hinton Agar, and Nutrient Agar as tests with high specificity and gold standard. In addition, to investigate the effect of added nanoparticles to bitumen samples, minimum inhibitory concentration (MIC) were determined by microdilution broth method with control sample to ensure the accuracy of the proceeds.

Results

As previously described, by Marshall method, two asphalt samples with specific grains were made. Samples consist of 4.5% bitumen (54g net bitumen) number 1 (Figure 1), an asphalt sample content of

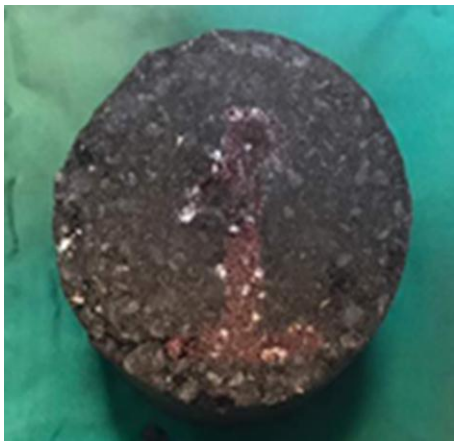

Figure 1. Normal asphalt samples.

Figure 3. Asphalt containing nanoclay.

nanocarbon single walled with 5.5% (3g) bitumen with number 2 (Figure 2) and an asphalt containing bentonite nanaclay with 5.5% (3g) bitumen defined with number 3 (Figure 3). At least four samples were produced in asphalt and bitumen laboratory in Iran University of Science and Technology. In addition, some old asphalt belongs to purview samples, as a sample 4 was collected and is shown in Fig4. From asphalt samples, the suspension was prepared in serum physiology that after cultured on Nutrient Agar, the old asphalt had more bacteria included *Bacillus* and *Pseudomonas*. The new asphalt had less bacteria. In nanoclay and nanocarbon asphalt, the rate of bacteria was significantly reduced than new and old asphalt so the reason of this can be the sealing mechanism of nanoparticles and inhibitory effect of them on the bacterial penetration into asphalt structure. In addition, nanoclay asphalt had more bacteria than nanocarbon that it shows nanocarbons have more efficacy. *Pseudomonas* and *Bacillus* are able to use asphalt materials as a

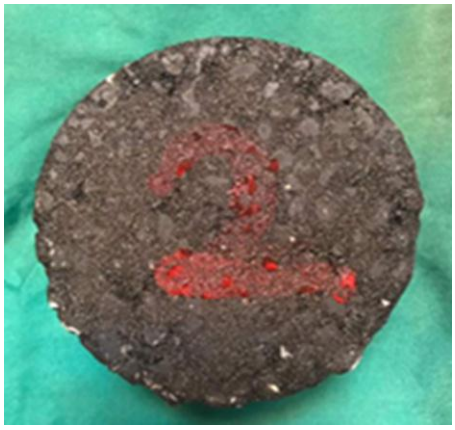

Figure 2. Asphalt containing nanocarbon.

substrate and decompose it. By the MIC method results, nanocarbon and nanoclay in 2% and 4% concentration was able to inhibit bacterial growth respectively. It shows using carbon nanoparticles at adequate concentration can cause more stability of asphalt.

Discussion

In the USA in 1963, bacterial genera such as *Pseudomonas*, *Bacillus* and *Chromobacterium* were isolated from asphalt as microbial agents, which decompose bitumen[13]. In addition, in the current study *Pseudomonas* and *Bacillus* were detected in asphalt samples. In a study in 1989, to survey effect of

Figure 4. Old collected asphalt.

microbial deterioration on the connection of asphalt component, routing, cracking, raveling, stripping and weathering, shoving and disintegration, all of these were common failures in pavement, which every year it costs, a lot to national economy of each country[6]. In

our country the costs of pavement and asphalt production are increasing, so according to our results it is better by identifying and fixing destructive factors such as bacteria, help to improve economy. In a study in 1989 by American society of microbiology, 7 aerobic, gram negative bacteria from an enriched culture medium from degraded asphalt isolated which the most isolated genera were *Pseudomonas*, *Acinetobacter*, *Alcaligenes*, *Flavimonas* and *Flavobacterium*. The strongest destructive bacteria was *Acinetobacter*, which destroyed aromatic compounds such as Naften[14]. In current study, the *Pseudomonas* and *Bacillus* were the main and only genera that found in asphalt. It is possible to be available different bacterial flora in other place. In a study in 2015 in Tabriz (East Azarbaijan province, northwest of Iran), demonstrated that soils contaminated with petroleum compounds, usually have bacterial species that break down aromatic compounds[15]. Therefore, it shows that there are different areas with different flora of soils, which can survey and prevent damage of asphalt and bitumen. The presence of clay and carbon nanoparticles in asphalt structures can increase the durability of the hot mix asphalts (HMA) and reduce the relevant economics costs. According to the destructive role of microorganisms in asphalt degradation that not investigated so far in Iran, this study has been innovated and it can increase the connection of road and transportation engineering sciences with medical sciences. Previous studies focused on petroleum compounds but the present study concentrated on the destructive role of microorganisms in asphalt and bitumen.

Conclusion

The old asphalt had more bacteria than other samples, which shows bacteria are one of the main factors in decomposition of asphalt by applying corrosion to bituminous hydrocarbons. Presence of clay and carbon nanoparticles in asphalt structures could increase the durability of the HMAs and reduce the relevant economics costs.

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