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The effects of motor oil on the growth of three aquatic macrophytes

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ABSTRACT

In this study, the effect of used motor oil on the growth rate of three different species of aquatic macrophytes was investigated for a three week period under laboratory conditions. Three treatments were used in pots: high oil, low oil, and a control, each with three replicate buckets (three pots per bucket). The relative growth rate (RGR) of the tested plants, *Potamogeton gramineus* L., *Myriophyllum spicatum* L., and *Ceratophyllum demersum* L., differed significantly between treatments ($p < 0.001$, one-way ANOVA). In the control treatment, *C. demersum*, *M. spicatum* and *P. gramineus* grew well and produced more lateral shoots than in the high and the low motor oil treatments. The longest shoot lengths were also greater for all three plants in the control than in the low and the high motor oil groups.

1. Introduction

Burned oil can negatively affect not only the soil but also the aquatic environment and the atmosphere. Therefore, used motor oil can be dangerous if not properly managed [1].

Lubricating oils mainly include 75 to 85% of base stock (i.e., crude oil) and performance-enhancing additives. The base stock consists of mineral oil, synthetic oil, or a blend of both. Mineral-based oil contains polynuclear aromatic hydrocarbons (PAH). It is well known that PAH such as benzo[*a*]pyrene are highly carcinogenic. However, oil additives can contain considerable quantities of hazardous chemical species for growth of aquatic macrophytes, such as zinc, magnesium, molybdenum, and phosphorus, sulphur and bromine compounds [2], which can be highly toxic to organisms [1].

One of the principal differences between new and used motor oil is the heavy metal concentration. Whereas used motor oil contains high concentrations of Pb, Zn and Ba, the concentrations of Fe, Cu, Al, Cr, and Ni are lower [3–6]. The nitrogen, sulphur, phosphorus and chloride contents of used motor oil are typically 0.05–0.18%, 0.22–0.55%, 80–32,000 $\mu g \cdot g^{-1}$ and approximately 35,000 $\mu g \cdot g^{-1}$, respectively [3–5]. Used motor oil also contains a high amount of heavy metals, such as Pb (13,000 $\mu g \cdot g^{-1}$), Zn (2500 $\mu g \cdot g^{-1}$), Cu (50 $\mu g \cdot g^{-1}$), Cr (20 $\mu g \cdot g^{-1}$), Ni (5 $\mu g \cdot g^{-1}$) and Cd (0.1 $\mu g \cdot g^{-1}$). Furthermore, significant amounts of other compounds have been reported, including Ca (4000 $\mu g \cdot g^{-1}$) Mg (1000 $\mu g \cdot g^{-1}$) and Na (600 $\mu g \cdot g^{-1}$) [3–6]. However, the PAH content of new motor oil is relatively low [7] but it increases during use depending on the time that the motor is in operation [8]. During use in a motor, the chemical structure of a lubricating oil is changed by oxidation, nitration, cracking of polymers and decomposition of organometallic compounds [9]. Used motor oil contains at least 50 $\mu g \cdot g^{-1}$ of PAH [7,10].

Invertebrates and fish have been commonly used to assess the potential damage of effluents and waste water, whereas photosynthetic organisms are restricted to estuarine regions [11–13]. However, there is an increasing interest in aquatic plants as potential indicators of water quality [14,15]. Aquatic macrophytes offer particular advantages as biomonitors of the environmental quality of aquatic ecosystems and there are many reasons why plants are better bioindicators than animals [16–19]. Because plants comprise the base of most food chains, the effects of toxic compounds released into water will become rapidly evident in organisms at higher trophic levels. Therefore, plants have a role in providing an early warning signal of possible contaminant effects on other trophic levels in aquatic environments. Macrophytes also directly respond to their local environment, in contrast to many mobile or top predators such as fish, whose diet might not be derived entirely from the aquatic food chain [20]. Although there are some studies about motor oil effect on the biomass of aquatic macrophytes [12,13,21], these studies have focused predominantly on free-floating, and semi aquatic plants. Therefore, the aim of this study was investigate whether motor oils affect the growth rate of submersed aquatic macrophytes due to the effect on water quality.

2. Materials and methods

Three species of macrophytes, *Potamogeton gramineus* L., *Myriophyllum spicatum* L., and *Ceratophyllum demersum* L., were

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collected with a grapnel from Lake Çalı, Turkey (41°12′N, 43°12′E) in June 2010. After being transported to the laboratory, plant material was separated from debris, and 10 cm shoots of each species were cut. The shoots of each species were contain 5–7 stem node. The wet weight of the shoots was recorded to calculate the initial dry weight. A total of 54, 10 cm length, individual *P. gramineus*, *M. spicatum*, and *C. demersum* shoots were inserted into 8 × 8 cm, 10-cm deep square plastic pots filled with lake sediment which consists of clay-sand gravel. Six pots were placed in 15-L capacity buckets and each bucket was filled with tap water which hard with high calcium and magnesium carbonate. After 5 days nursery culture, unhealthy plants were removed from buckets and the experiment proper was started with the total of 9 buckets and 27 pots; three pot treatments (low motor oil, high motor oil and a control), each with three replicate buckets. The concentration of PAH in run-off water from roads carrying high-density traffic varies from 6–24 $mg\cdot l^{-1}$ and that from residential zones varies between 0.2 and 7.5 $mg\cdot l^{-1}$ [22]. To create the low motor oil treatment, used motor oil collected from a car mechanical workshop was placed on the surface of the water at 1 $mg\cdot l^{-1}$ without mixing with the substratum and avoiding direct contact with plants. In the high motor oil treatment, 15 $mg\cdot l^{-1}$ of used motor oil was added to the water using by same method as the low treatment. Containers were maintained in the laboratory under a 12:12 h light:dark cycle. The light irradiation at the water surface level was 90 μmol PAR (Photosynthetically Active Radiation) m^{-2} s^{-1}. The water temperature was maintained at 15 °C during the experiment. Samples for physico-chemical analyses were taken immediately after the buckets were set up and again at the end of the experiment. In each bucket, dissolved oxygen (DO), conductivity, pH and temperature were measured with a WTW Oxi 197i oxygen meter (Weilheim, Germany), a WTW cond 315i/set meter (Weilheim, Germany), a WTW 315i/set pH meter (Weilheim, Germany), and a thermometer, respectively. Light was measured with a Macam Quantum Radiometer/Photometer Q101 (Macam Photometers Ltd., Livingston, Scotland) fitted with an underwater probe.

In the laboratory, NH_4–N, NO_3–N and Soluble Reactive Phosphorus (SRP) were analysed according to American Public Health Association (APHA) methods [23]. Chlorophyll *a* (chl *a*) was extracted in acetone and its concentration was calculated from the absorbance reading at 663 nm [24]. The content of heavy metals (Pb, Zn, Cd, Cr) and aromatic hydrocarbons, such as benzene, toluene and xylene, in the used motor oil were determined according to US EPA Method 3050A [25] and US EPA Method 8020B [26], respectively.

After 21 days, the plants were harvested and each plant species was dried to constant weight in an oven at approximately 70 °C. The Relative Growth Rate (RGR) of each plant was calculated based on the dry weight as

$$RGR = [\text{In(final dry wt.)}–\text{In(initial dry wt.)}]/\text{duration of the experiment}$$

To obtain the initial dry weight, a functional approach was used. The wet weight and dry weight ratio of each plant shoot was calculated at the end of the experiment and the mean values were used to extrapolate the initial dry weight. For statistical analyses, a one-way analysis of variance (ANOVA) was used.

3. Results and discussion

Used motor oil contained higher concentrations of Pb (354.3 $mg\cdot l^{-1}$) and Zn (810 $mg\cdot l^{-1}$), but lower concentrations of Cr (6.15 $mg\cdot l^{-1}$) and Cd (2.17 $mg\cdot l^{-1}$) (Table 1). The nitrogen and phosphorus concentrations of the tap water that was used as the liquid medium were high; therefore, the tap water supplied some nitrogen and phosphorus for plant growth (Table 2). In the control treatment, *C. demersum*, *M. spicatum* and *P. gramineus* grew well and produced more lateral shoots than in the high and the low motor oil treatments (Table 3). The longest shoot lengths were also greater for all three plants in the control than in the low and the high motor oil groups. As an aquatic rhizomatous herbs, *M. spicatum* and *P. gramineus* did not produce rhizomes in any of the experimental group. The RGR of *C. demersum*, *M. spicatum* and *P. gramineus* differed significantly between treatments ($p < 0.001$, $p < 0.001$ and $p < 0.001$, respectively, using one-way ANOVA) (Fig. 1).

Table 1
Concentrations of heavy metals and hydrocarbons in used motor oil. Values shown are means (n = 3) with standard deviations in parentheses.

Metal/hydrocarbon	$mg\cdot l^{-1}$
Pb	354.3 (86.45)
Zn	810 (215.01)
Cr	6.15 (1.67)
Cd	2.17 (0.75)
Benzene	12.8 (1.23)
Toluene	208 (77.31)
Xylene	393 (154.69)

The heavy metal content of used motor oil is very important because many heavy metals are potentially dangerous to living organisms. In this study, the observed Pb and Zn concentrations were much lower than in other types of oils that have been investigated [6,4]. These changes can likely be attributed to changes in the formulation of unleaded gasoline; however, both metals remain potentially toxic.

Used motor oil accumulates not only heavy metals but also aromatic hydrocarbons that are present in insignificant amounts in unused oil. These particles principally originate from atmospheric dust, metal oxides and combustion products. In this study, the hydrocarbons benzene, toluene and xylene were analysed. The results indicated that the used motor oil was enriched in hydrocarbons at similar concentrations to those found in used motor oil by other research groups [27].

Motor oil spills generally occur on land, and this oil can be rapidly transported to the aquatic environment by rain and run-off water [1]. The primary effect of hydrocarbons in aquatic environments is a change in the composition of communities of microorganisms [28]. Generally, hydrocarbons in the aquatic environment stimulate an increase in yeast and bacteria that can degrade them; hydrocarbons also suppress the growth of phytoplankton species, which are pollution-sensitive. Petroleum hydrocarbons inhibit the growth, the photosynthetic activity and the respiration of algae [29,30]. However, in the present study, the chl *a* concentration increased with increasing amounts of motor oil (Table 2, $p < 0.005$ for all tested plants during week 3). Hydrocarbons can stimulate microbial growth at low concentrations [1] and this might explain why higher chl *a* concentrations were found in the low motor oil treatment than in the controls. However, several physico-chemical factors clearly determine the effects of hydrocarbons on algae. For example, the presence of bacteria that can degrade hydrocarbons can limit the toxic effects of hydrocarbons on photosynthetic organisms [31,32], which might explain why the chl *a* concentration increased with an increase in the motor oil concentration in this study.

In contrast to the chl *a* concentration, the growth rate of all three macrophyte species decreased with an increase in the motor oil concentration. Some aquatic plants are more sensitive to toxicants (e.g., atrazine) than other organisms [33]. Taraldsen and Norberg-King [34] and Swanson et al. [35] have shown that free-floating macrophytes are less susceptible to several toxins than rooted macrophytes. Similarly, many studies that have measured metal accumulation by plants have shown that rooted submerged plants accumulate higher concentrations of metals than free-floating species [36,37]. From the three macrophyte species used in this study, two of them, *M. spicatum* and *P. gramineus*, are rooted submerged plants, but *C. demersum* is a rootless submerged species. According to the study of Matache et al. [38] *Ceratophyllum* concentrates higher amount of heavy metals compared *Potamogeton* species, being a better accumulater. Samecka-Cymerman and Kempers [39] found higher concentration of cadmium and lead in *Myriophyllum*