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Review

A review of germination and early growth as a proxy for plant fitness under petrogenic contamination — knowledge gaps and recommendations

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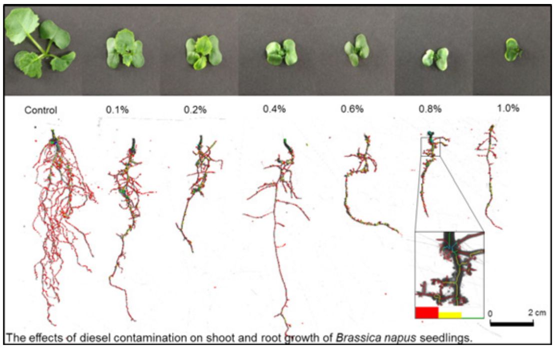
HIGHLIGHTS

- Petrogenic contamination effects on germination and seedling stages are documented.
- The use of root imaging software is proposed for ecotoxicological assessments.
- Key traits are suggested to inform selection criteria of appropriate plant species.
- Recommendations are made to improve germination protocols.

GRAPHICAL ABSTRACT



The effects of diesel contamination on shoot and root growth of *Brassica napus* seedlings.

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ABSTRACT

Germination—an important stage in the life cycle of plants—is susceptible to the presence of soil contaminants. Since the early 1990s, the use of germination tests to screen multiple plant species to select candidates for phytoremediation has received much attention. This is due to its inexpensive methodology and fast assessment relative to greenhouse or field growth studies. Surprisingly, no comprehensive synthesis is available of these studies in the scientific literature. As more plant species are added to phytoremediation databases, it is important to encapsulate the knowledge thus far and revise protocols. In this review, we have summarised previously-documented effects of petroleum hydrocarbons on germination and seedling growth. The methods and materials of previous studies are presented in tabulated form. Common practice includes the use of cellulose acetate filter paper, plastic Petri dishes, and low numbers of seeds and replicates. A general bias was observed for the screening of cultivated crops as opposed to native species, even though the latter may be better suited to site conditions. The relevance of germination studies as important ecotoxicological tools is highlighted with the proposed use of root imaging software. Screening of novel plant species, particularly natives, is recommended with selection focussed on (i) species phylogeny, (ii) plant morphological and functional traits, and (iii) tolerance towards harsh environmental stresses. Recommendations for standardised protocols for germination and early growth monitoring are made in order to improve the robustness of statistical modelling and species selection in future phytoremediation evaluations and field programs.

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Contents

1. Introduction

Anthropogenic releases of petrochemicals contaminate terrestrial ecosystems with high-molecular-weight organic compounds of a carcinogenic and/or mutagenic nature (Banks and Schultz, 2005; Corgié et al., 2004; Dong and Lee, 2009; Maila and Cloete, 2002). Spillage or loss of containment events commonly occur as a result of activities associated with the extraction, production and transportation of oil for both manufacturing industries and agricultural operations. Subsequent clean-up expenses for contaminated sites can run into billions of dollars. For example, in the USA, the largest producer of oil, it is estimated that the clean-up of all current hydrocarbon-contaminated sites will cost approximately US$1 trillion (Kuiper et al., 2004; Stroud et al., 2007). In soils 'aged' with hydrocarbon contaminants, the residence time of recalcitrant fractions and their breakdown products can exceed multiple decades. For example, Rezek et al. (2008) experimented with remediation of soil contaminated as long ago as World War II (1939–1945) and found that even after 50 + years, the concentrations of most polycyclic aromatic hydrocarbons (PAHs) were still high and they had not degraded significantly after 18 months of vegetation or fertilisation amendments. Such long-term persistence presents a high risk of contaminants leaching into groundwater and their potential bioaccumulation in the food chain; this is widely discussed in the literature (Collins et al., 2006; Fujikawa et al., 1993; Gao and Zhu, 2004; McCready et al., 2000; Meador et al., 1995; Siddiqui and Adams, 2002; Simonich and Hites, 1995). As a result, the United States Environmental Protection Agency (USEPA) has registered 16 such PAHs as hazardous priority contaminants (Lamichhane et al., 2016).

As physical methods of remediation such as incineration and thermal desorption are unacceptable both on an environmental and economical, scaleable basis (Henner et al., 1999; Joner and Leyval, 2001; Smith et al., 2006), biologically-assisted remediation, i.e. remediation by soil microorganisms in the absence of plants, has been successfully employed. Traditionally, bioremediation techniques have been studied in conjunction with fertiliser supplements to ensure C:N:P ratios favour high microbial activity (Atlas, 1991; Dibble and Bartha, 1979; Walworth and Reynolds, 1995). In general, a molar ratio of 100:10:1 is recommended and used in most studies (Leys et al., 2005). However, hydrocarbon spills also cause ecological disturbance of the soil indigenous micro-biota (Megharaj et al., 2000) and fertiliser additions rarely aid recuperation by the microbial biota to the pre-spill state, as it is mostly hydrocarbon degrader communities that persist. Indeed, evidence suggests that increased use of fertilisers can cause ammonia or nitrite toxicity to microorganisms (Tibbett et al., 2011).

In the past two decades, phytoremediation (the use of plants to degrade contaminants of all types, e.g. organic, heavy metals) as a 'green liver' concept has been extensively investigated in relation to the degradation of hydrocarbon contaminants (Sandermann, 1994). Phytoremediation garners high public acceptance due to its apparently sustainable and ecologically-holistic approach. Moreover, numerous studies have reported greater degradation of contaminants in the presence of plants than in microbe-only controls (Aprill and Sims, 1990; Binet et al., 2000; Gaskin and Bentham, 2010; Günther et al., 1996; Meng et al., 2011; Miya and Firestone, 2001; Peng et al., 2009; Reilley et al., 1996). However, understanding the underlying mechanisms involved in the breakdown phases of a natural phyto-remediator system is slow owing to the complexity of the multiple mechanisms that plants can employ to remove the hydrocarbons (Table 1). It is also important to point out that some studies have reported that the growth of test plants in hydrocarbon-contaminated soil failed to catalyse efficient degradation compared with unplanted controls (e.g. Fang et al., 2001; Ferro et al., 1997; Olson et al., 2007b; Watkins et al., 1994). Plant species may differ in their tolerance of hydrocarbon contaminants; therefore, it is essential to screen more plant species for hydrocarbon tolerance to identify the most tolerant species and those best-suited to specific situations as there is huge variability in soil type, contaminant history, climatic conditions and nutritional status of contaminated sites. This may increase the likelihood of selecting species effective in causing biodegradation.

Screening of plant species for hydrocarbon tolerance, based on growth characteristics, requires months of plant development (Marques et al., 2010); maintenance of a large number of species in a replicated experiment over this time is expensive (Kirk et al., 2002). Associated procedures such as solvent extraction of contaminated soils and gas chromatography–mass spectrometry (GCMS) analyses of the extracts add further to the costs. To fast-track screening and reduce operational expenses, many authors have supported the use of germination screening tests (Maila and Cloete, 2002; Smith et al., 2006). Also, the most effective approach to establishing vegetation on disturbed lands, such as contaminated sites, is direct seeding, as it ensures better survival and is cheaper than alternative transplanting techniques (Merritt and Dixon, 2011; Palmerlee and Young, 2010). Even so, high germination rates are desirable to reduce seed costs. To our knowledge, this review is the first attempt to consolidate the present knowledge on seed germination and early growth in hydrocarbon-contaminated soils.