Marine Macroalgae as a Source for Osmoprotection for 
*Escherichia coli*

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Abstract. At elevated osmolarity of the mineral medium M63, marine macroalgae constitute important osmoprotectants and nutrients sources for *Escherichia coli*. Growth of bacterial population (16 strains) was improved by supplementing M63 salts medium with either aqueous or ethanolic algal extracts obtained from *Ascophyllum nodosum*, *Fucus serratus*, *Enteromorpha ramulosa*, *Ulva lactuca*, and *Palmaria palmata*. In their presence, growth was still observed even at 1.02 M NaCl. Furthermore, the *E. coli* ZB400 growth in presence of whole macroalgae thalli in M63/0.85 M NaCl reached its maximum within 24 h (5 × 10⁷ – 5 × 10⁸ colony-forming units [CFU] per milliliter). In the presence of *A. nodosum*, bacterial growth was inhibited. In the same experimental conditions, ethanolic extracts improved *E. coli* growth significantly, because the yield reached 10¹¹ CFU per milliliter. *Ulva lactuca* and *P. palmata* allowed the better growth. The Dragendorff-positive compounds extracted from bacterial cells growing on each ethanolic extract exhibited an osmoprotective effect as proved by a disk-diffusion assay. On the other hand, the -onium compounds (quaternary ammonium [betaines] and tertiary sulphonium) and total free amino acid contents of *U. lactuca* ethanolic extracts were higher than in others. Fucaceae extracts demonstrated especially high protein content. Algal extracts constitute not only an appreciable osmoprotection source for *E. coli* but also nutrient sources.

Introduction

Survival of enteric pathogens in coastal environments has been established [21]. Pathogenic bacteria (*Vibrio cholerae*, *Salmonella* spp., and *Escherichia coli*) proba-

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bly enter into dormancy, during which they remain potentially virulent [11, 21, 22, 39]. Thus, microbial pollution of the littoral presents a serious public health hazard, especially via the alimentary chain. Survival of \textit{E. coli} is affected by, among others, oligotrophy and salinity [1, 6, 23, 30]. Longer survival and growth of coliforms were observed in the presence of marine sediments, compared to unsupplemented seawater. Organic matter in the sediments constitutes a carbon and osmoprotective source [7, 18–20]. Osmoprotectants, when supplied in the medium, allow \textit{E. coli} to overcome deleterious effects of osmotic saline stress [13].

It is well established that -onium compounds (quaternary ammonium [betaines] and tertiary sulphonium) [38] are involved in osmoregulatory mechanisms of \textit{E. coli} [8, 12, 25]. Such compounds are widespread in nature and present within organisms exposed to saline environments [2]. As such, marine macroalgae consistently yield either betaines or their sulphonio analogues [3, 4]. Either one or both of these classes of compounds are a constant feature of marine macroalgae. Glycine betaine (GB), \(\gamma\)-butyrobetaine (\(\gamma\)BB), proline betaine, and dimethylsulphoniopropionate (DMSP) have a common and widespread distribution. They likely contribute to alleviating salt or water stress. The possible role of DMSP in the maintenance of osmotic balance has been pointed out in \textit{Ulva lactuca}, \textit{Enteromorpha intestinalis}, and \textit{Polysiphonia lanosa} [14–16, 33]. Osmoregulatory activities could be intense within macroalgae occupying the intertidal zones and estuaries and thus affected by rapid changes in salinity that are due to the interactions of tides, rainfall, and evaporation. Thus, osmoprotectants may become released to the marine environment, which is due to excretion, leakage, or death of macroalgae.

High quantities of macroalgae are driven ashore on Brittany coasts, particularly during summertime; their decomposition may favor the bacterial growth and pollution. Recently, Flatau et al. [17] suggested that the survival of \textit{E. coli} in marine environments might depend on osmoprotective compounds released into seawater by phytoplankton or halophilic phanerogams. However, the relation between marine macroalgae and \textit{E. coli} was not clearly demonstrated until now. Previous observations (M. Ghoul, 1990, doctoral thesis, University of Rennes I, France) demonstrated that \textit{E. coli} accumulated several Dragendorff-positive compounds from each of the following macroalgae: \textit{Ascophyllum nodosum}, \textit{Fucus serratus}, \textit{Enteromorpha ramulosa}, \textit{U. lactuca}, and \textit{Palmaria palmata}. We report here that, under laboratory conditions, algal extracts or whole thalli of the above macroalgae markedly restore \textit{E. coli} growth in high osmolarity mineral medium.

\textbf{Materials and Methods}

\textbf{Algal Materials}

The following common macroalgae of Brittany coasts (Northwestern France) were used: \textit{A. nodosum} (L.) Le Jolis, \textit{F. serratus} L. (Fucaceae), \textit{E. ramulosa} (Smith) Hooker, \textit{U. lactuca} L. (Ulvaceae), and \textit{P. palmata} (L.) Kuntze (Palmariaceae). They were collected from the intertidal region of Tregastel (Brittany) during spring and summer. After collection, thalli were washed thoroughly with cold, natural seawater before use.