INTERNATIONAL COUNCIL FOR
THE EXPLORATION OF THE SEA

C.M. 1981/E : 56
Marine Environmental
Quality Committee

BIODEGRADABILITY IN THE MARINE ENVIRONMENT. I. THE CHEMOSTAT AS A MODEL SYSTEM FOR THE AQUATIC ENVIRONMENT.

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Summary
A test method is described for measuring biodegradability in the aquatic marine environment. On the basis of the characteristic properties of the sea the chemostat has been chosen as the model system. In the chemostat test the shortest possible residence time for a test compound is established. On the basis of this residence time a turn-over time in the natural environment is estimated.

Introduction

As man continues to develop and utilize new organic chemicals, the potential for environmental pollution increases accordingly. Once a chemical has entered the environment its eventual transport to the aquatic environment becomes an obvious possibility.
At the moment more than 50 000 chemicals are produced commercially and each year about 100 new chemicals are added to this list.

Before a chemical is manufactured commercially, a hazard assessment has to be made to establish the environmental risk of this chemical. Besides, in case of an inadmissible risk, a risk benefit analysis could be performed. Two key questions in the environmental hazard assessment are:
. what is the fate of the chemical in the environment,
. which effects may be expected.
A thorough knowledge of the fate of a chemical is required and particularly with respect to its ultimate removal from the environment by biological or chemical (12) degradation.

Micro-organisms are commonly believed to play the most important role in determining the fate of chemical substances in aquatic systems. In the marine environment most of the organic matter produced photosynthetically in the surface waters is decomposed in the upper few hundred meters (9) especially by heterotrophic bacteria (1). In coastal and open waters 90 % of the measurable heterotrophic uptake occurred in the fraction less than 1 µm in size or was associated with bacteria on particles (1,9).

Literature studies reveal few research concerning the rates and products of microbial degradation in the aquatic environment of most of the commonly used chemicals. The greater part of the research has been pointed at

the metabolic pathways by which certain chemically pure substances were transformed by pure or enriched cultures in a well defined medium (3, 10, 13).
Although a large number of bio-degradability testing methods are available (4), at the moment not yet one suitable method for the measuring of the aquatic environmental degradation exists; perhaps because of the great variety in aquatic compartments (fresh versus marine, estuaries, coastal zones, oceans, deep sea).
Particularly considering the marine environment no specific methods have been developped. One has to rely on existing methods for the fresh water environment.

This study has been aimed at the development of a test method for the prediction of the bio-degradability potential of organic chemicals in the marine environment.

Choice of the test system

To predict the fate of xenobiotic substances in the marine environment a model system has been developped resembling the sea. Important characteristic properties of the marine environment are:

- a continuous input of organic substances by means of the primary production of phytoplankton,
- a continuous decomposition of organic material by mixed populations of heterotrophic micro-organisms,
- a dynamic equilibrium during this continuous carbon cycle,
- a wide range of turn-over times for all kinds of compounds in various areas (6).

Combining these characteristics a continuous flow culture according to the chemostat principle (16) has been chosen as model system for the marine environment.

The primary production is simulated by the input of a large number of fairly bio-degradable organic chemicals, in relatively high concentrations regarding to the natural marine environment, but in about the same relative amount. In this way a relatively higher biomass is obtained with relatively higher bio-degradation rates than in natural seawater. The organic substrates together constitute as carbon and energy source the growth limiting factor for the heterotrophic micro-organisms.

The natural environment contains a lot of heterogeneous assemblies of microbes and the promotion of mixed cultures may give better results, especially for xenobiotic chemicals (14). The continuous input of a large number of organic substances promotes the maintenance of a well mixed community of micro-organisms in the chemostat and may stimulate cometabolic transformations (8). Cometabolism is the metabolism by a micro-organism of a chemical which this species cannot use as a nutrient or energy source (7, 11). Consequences of cometabolism are a slow microbial transformation rate and the accumulation of structurally similar products. However, the combination of a cometabolic step and mixed cultures may still lead to complete degradation, in which reaction sequence the cometabolic step(s) is(are) likely to be rate limiting.

In a chemostat just as in the sea a dynamic equilibrium arises which cannot be disturbed easily because this system is self-stabilizing.

The turn-over time of a chemical is the time needed for the complete degradation of this chemical. The turn-over time in the sea can be compared with the residence time in the chemostat. The residence time is the mean time during which the substrates and the micro-organisms stay in the culture vessel and during which degradation by the community of microbes may

occur. In the chemostat a wide range of residence times is possible. A short applicable residence time for a chemical does also mean a short turn-over time.

The study for the microbiological fate of a xenobiotic compound is performed by adding the xenobiotic to the nutrient solution in minor, non-toxic, but analytically suitable concentrations. The degradation is established as function of the residence time. Figure 1 gives a degradation curve. As characteristic data from this curve the range of residence times (T1 to T2) for the transition from 0 to 100 % degradation is defined.

Interpretation of test results.

The degradation of organic compounds in the natural aquatic compartments can be estimated by assuming that the decomposition of the organic substances equals the production of these compounds by the primary production, because the standing crop of organic carbon in the oceans is in a steady state. If the biomass present is known the degradation rate, the amount of dissolved organic carbon degraded per unit of time and biomass, can be calculated. The turn-over of the organic substances of the nutrient solution in the chemostat is calculated for the highest possible degradation rate (point A in figure 1), situated in the characteristic residence time range T1 - T2. From the performed experiments (2) this range may be estimated at 0.75 to 1.0 h. Table I shows the degradation rates for organic carbon in the various natural aquatic compartments and the chemostat. From the data in table I a theoretical ratio between the degradation rates of organic compounds in the chemostat and in the other aquatic compartments can be calculated (table II).

For glucose some literature data are available of turn-over times in various aquatic compartments (6, 15) and for the chemostat the minimum necessary residence time for complete degradation may be estimated at 0.75 h (2). A large range is seen in the turn-over data for glucose, but in comparison with these experimental data the ratio between chemostat and "naturally" measured data is about the same as theoretically calculated (table II).

Conclusions

It seems possible to predict residence times or turn-over times in aquatic marine compartments on the basis of the data obtained in chemostat experiments.

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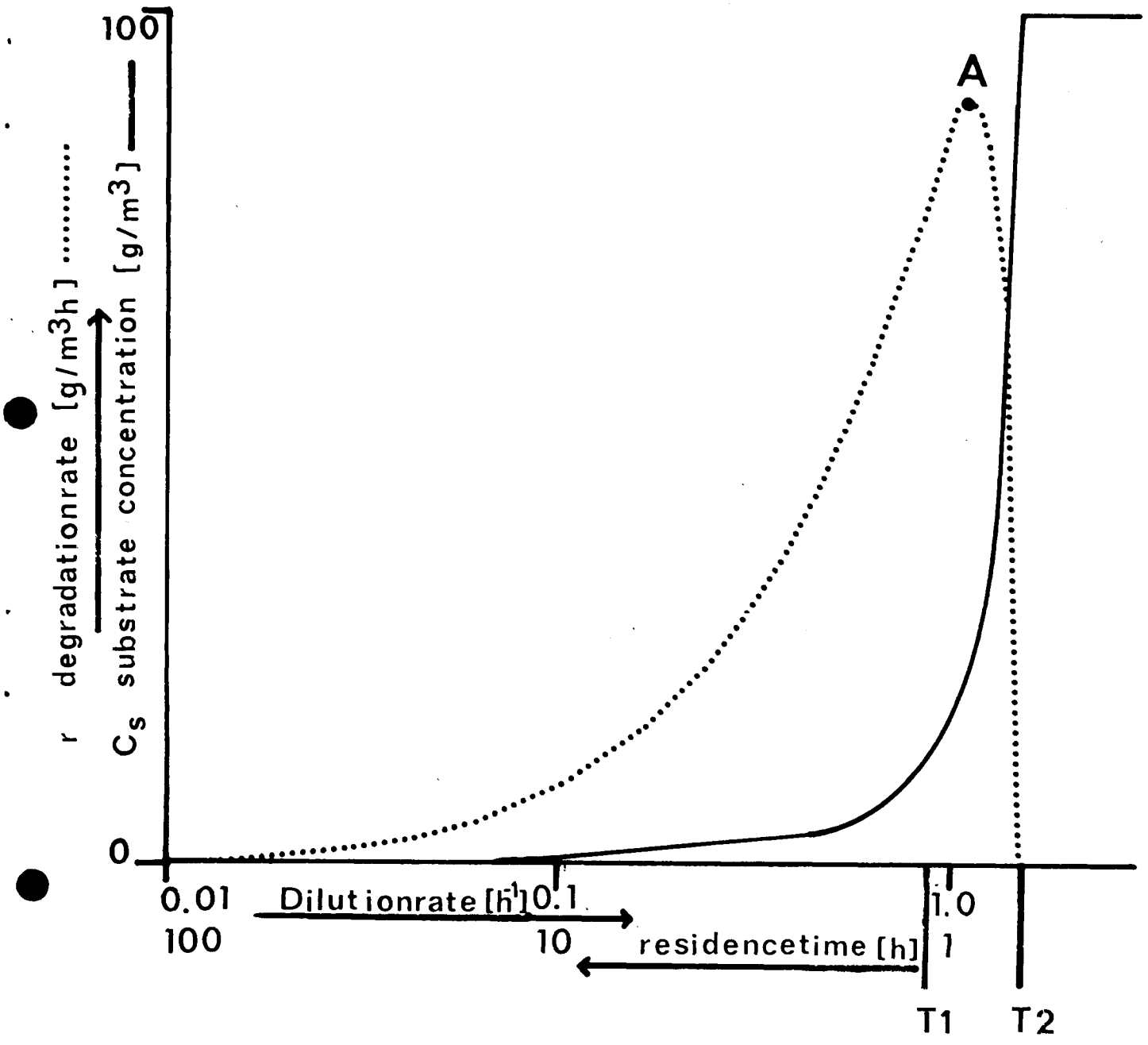

FIGURE 1. Concentration and degradationrate of a substrate in the chemostat as a function of the residencetime.

TABLE I. Degradationrate of organic carbon in various aquatic compartments.

	primary production gC/m^2a	depth[a] m	degrad. rate gC/m^3a	biomass[b] mg/m^3	degrad. rate $gC/g.a$
oceans	50	200	0.25	5 - 50	5 - 50
estuaria coastal zones	100 - 500	20	5 - 25	10 - 100	50 - 2500
chemostat			10^6	10^5	10^4

notes
a. depth in which the degradationprocesses take place
b. biomass as dry weight

TABLE II. Ratio of the degradationrates (A) and experimental data for glucose (B).

	(A) ratio degr.rate chemostat - compartment	(B) glucose degrad.rate experimental data [h] median	range
ocean	200 - 2000	1000[a]	200 - 6000[a]
estuaria coastal zones	4 - 200	20[a]	1 - 500[a]
chemostat	1	0.75[b]	

notes
a. turnovertime - literature data (6, 15)
b. residencetime (2)