Hydrolysis:
Important test to care an environment

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Definition

Hydrolysis is a chemical reaction during which molecules of water (H_2O) is split into two ions: hydrogen cations (H^+) and hydroxide anions (OH^-) and attached to two parts of a molecule.

Introduction

Hydrolysis is the most common reaction controlling abiotic degradation and thus it is one of the most degradation pathways of the chemical in an environment. Chemicals, encountering the surface waters, through various routes e.g., direct application, drainage, run-off, industrial waste disposal, domestic or agricultural effluent, and spray drift, may be transformed via hydrolysis, oxidation, photochemical and microbial processes. Hydrolysis is especially important for the materials with low biodegradability that may persist in the environment after use and disposal. With a better ability to transform and degrade, chemicals in the atmosphere generally have less potential for persisting as an environmental pollutant.

Test application

The test is generally suitable for technical grade products and not suggested for formulated products, as the formulation ingredients may influence the hydrolysis process. The test applies to slightly volatile and non-volatile compounds having a sufficient solubility in water. The test should not be applied to compounds that are highly volatile from water. The test may be carried out with labelled or non-labelled test compounds. Usually, the ^{14}C labelled test compound is preferred for studying the pathway of hydrolysis and the establishment of mass balance. Wherever possible, the label should be positioned in the most stable part of the molecule. If the test compound contains one ring, labelling on this ring is required. If the test compound has two or more rings, separate studies may be needed to assess the fate of each labelled ring and to get relevant information on the formation of hydrolysis products[1,2,3,4].

Precautions requirement

Other than hydrolysis, the chemical can be transformed through oxidation, photodegradation, and microbial process while encountering water. To prevent this reaction and to measure the hydrolysis reaction, care should be taken as mentioned below[1,2,3,4]:

Oxidation

purge all the solutions using an inert gas (e.g., Nitrogen) to remove oxygen

Photodegradation

test should be carried out in the dark condition

Microbial process

sterilise all the glassware, solutions and test the sterility.

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Test Design

The test is performed at three pH values which are normally found in the environment (pH 4 – 9). The test can be performed for a hydrolytically unstable compound for the physiological purpose at pH 1.2 and temperature 37 °C, if required.

The test is designed to a tiered approach as mentioned below:

Tier-I (Preliminary Test)

The test is performed using one concentration (< 10^{-2} M or half-saturation) in three buffer solution of pH 4.0, 7.0 and 9.0 at temperature 50 °C for five days. If hydrolysis is observed <10% after five days, the test compound is considered hydrolytically stable and higher tier test is not required.

Tier-II (Hydrolysis Kinetic)

The test is performed in the sterilized buffer solution treated with the test compound at the pH values at which the compound was found unstable during the preliminary test. Required buffered solutions of the compound are incubated in a constant temperature water bath at the three selected temperature between 10 to 70 °C. Minimum six spaced data points between 10 % and 90 % hydrolysis of the reaction solution are analyzed at an appropriate time interval to test first-order behavior. The test is run until 90% hydrolysis of the compound is observed or for 30 days, whichever comes first.

Tier-III (Identification of hydrolysis products)

Any major hydrolysis product(s), which is forming > 10% of the applied dose is identified using a suitable analytical method.

End Points

Hydrolytic stability

Half-life calculation

Hydrolysis rate constant

Identification of major transformation products

JRF has vast experience and expertise to undertake the test for ^{12}C and/or ^{14}C test compounds following OECD, EPA, EC and JMAFF guidelines.

References

1. OECD, 2004: The Organisation for Economic Co-operation and Development (OECD) Guidelines for the Testing of Chemicals, OECD 111, Hydrolysis as a function of pH, adopted by the Council on April 13, 2004.
2. EC, 2008, COUNCIL REGULATION (EC) No 440/2008 of 30 May 2008, laying down test methods pursuant to Regulation (EC) No 1907/2006 of the European Parliament and of the Council on the Registration, Evaluation, Authorisation and Restriction of Chemicals (REACH), C.7. Degradation — Abiotic Degradation: Hydrolysis as a Function of pH, Official Journal No L 54, 01/03/2016.
3. Japanese Ministry of Agriculture, Forestry and Fisheries (JMAFF). Testing Guidelines for Physico-chemical properties study for Japanese Regulatory, Notification No.12-8147 "Data requirement for supporting registration of pesticides", Hydrolysis Studies, 2-9-13.
4. US EPA, 2008: The United States Environmental Protection Agency (EPA), fate, transport and transformation test guidelines, OPPTS 835.2120, Hydrolysis (EPA 712-C-08-012), October 2008.

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Specialised in organic chemistry, Research Officer in the Environmental Fate and Metabolism (EFM).

He has a good experience of conducting E-Fate studies like Carbon and Nitrogen Transformation Tests, Hydrolysis, Photolysis, Ready Biodegradability, Method development and validation. He is actively involved in research validations. He has professional experience of more than 13 years, including academic research, Synthesis R&D, medicinal plant extraction & isolation, analytical instrumentation, and CRO industry.

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For over 20 years in industry, Arun has a vivid experience, viz., leadership, training and designing, conducting and reporting of the environmental fate studies. He is adept in conducting a higher tier metabolism, biodegradation, sorption studies and identifying key metabolites using 12C and 14C test compounds, as per the GLP requirement, for the registration of a compound under OECD, EPA, and EU. He has participated in the training conducted at the Bhabha Atomic Research Centre (BARC) on the use and handling of radioactive substances in the field of research and is a certified Radiological Safety Officer.

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