Ascertaining the Biodegradability and Aquatic Toxicity of Biolubri Greasekote 100 Environmental Acceptable Lubricant (EAL) in Accordance with OECD 301F and OECD 201 Test Methods

Nelson Cheng[1] PhD (Honoris Causa) Patrick Moe[2] BSc, Grad Dip, MSc,
Magna International Pte. Ltd, 10H Enterprise Road, Singapore 629834.

Dr. Benjamin Valdez Salas[3], Dr. Michael Schorr Weiner[4]
Instituto de Ingeniería Universidad Automonous Baja California (UABC)

Abstract

The primary purpose of this article entails the details of Biolubri Greasekote 100 an Environmental Acceptable Lubricant (EAL) and the tests involved to meet environmental requirements of Swedish Standard SS 155470 [1] and Test Procedure for Biodegradability and Toxicity Test.

Keywords

Swedish Standard SS 155470, OECD 301F Biodegradability Test and OECD 302 Toxicity Test, Environmental Acceptable Lubricant (EAL) Vessel General Permit (VGP).

Background

The issue of marine pollution has gained international scrutiny in the wake of rising emission levels with growth in sea freights and the commercial cruise market.

Biolubri Greasekote 100 EAL lubricating grease is especially suited for applications where there is a high potential for lubricating products to be lost into the natural environment such as wire-rope lubricants, controllable pitch propeller thrusters paddle wheel propulsion, Stern tubes, thruster bearings, stabilizers, rudder bearings (excluding head bearing), azimuth thrusters, propulsion pod lubrication, submersible recovery and others that may be normally submerged Mechanical equipment subject to immersion including dredges and grabs.[2]

The name environmentally acceptable lubricants (EAL) is used to describe bio-based lubricants that have successfully met the standards for biodegradability, toxicity and bioaccumulation potential. [3] They are likely to have little or no impact on the aquatic environment compared to conventional lubricants. Lubricants that are expected to have desirable environmental qualities, they will have a minimal impact on the natural environment.

Regulatory enforcement is expected to be stepped up to minimize ship source pollution in efforts to reduce pollution levels at ports and at sea. A major source of ship source pollution is from the stern tube, a hollow tube in the hull of the ship which allows the propeller shaft to connect to the engine [4].

The oil-lubricated stern tube generates high levels of pollution from leakage through stern tube seals, leaking an estimated 80 million litres of oil into the ocean per year. Cumulative pollution at this rate compounds chemical toxicity, negative ecological changes and clean-up costs, prompting the US government to enforce regulations on commercial vessels in US waters. US EPA 2013 Vessel General Permit (VGP) was enforced to better address oil pollution.

VGP (Vessel General Permit) requirements make it mandatory for lubricants to meet stringent testing specifications for biodegradability, minimally-toxicity and bioaccumulation standards in order to be classified as Environmentally Acceptable Lubricants (EAL). [4,5]

All vessels constructed on or after 19 December 2013 must use an environmentally acceptable lubricant in all oil-to-sea interfaces." and it continues with "For all vessels built before 19 December 2013, unless technically infeasible, [4] owners/operators must use an environmentally accepted lubricant in all oil to sea interfaces."

Environmentally acceptable lubricants are defined according to the document as biodegradable, non-toxic and not bio-accumulative. Although this is a US legislation it will undoubtedly affect Europe as well. For one thing the VGP opens up new opportunities for European manufacturers and marketers since it will affect any vessel owner or operator whose business depends on travelling through American waters.

Under VGP regulations, vessels are required to use EAL if they fall into the following categories: Commercial vessels greater than 79 feet in length, Vessels operating in a capacity as a means of transportation Vessels with discharges incidental to their operations, Vessels which enter within three nautical miles of US coastlines and inland waters [6,7,8]

In view of VGP regulations, vessel owners and ship maintenance businesses must ensure that lubricants used on vessels meet EAL requirements. EALs are formulated to ensure that their impact on the environment is significantly reduced when compared to that of traditional hydrocarbon-based lubricants.

Introduction

Magna International Pte Ltd together with Universidad Autonomous Baja California (UABC) on-going pursuit of sustainability and growing concern for marine environment has led both the company and institution to increasingly consider the use of bio-based lubricants as replacement for conventional hydrocarbon lubricants. Bio-based greases degrade into simple substances not harmful to the ecosystem and making use on renewable raw materials.

The development of Biolubri Greasekote 100 was one of the results of the on-going research between Magna International and Universidad Autonomous Baja California (UABC).

Magna Biolubri Greasekote 100 is specially developed for the lubrication of marine, oil rigs and equipment onboard ocean-going vessels, forestry machinery, construction vehicles, rail curve, rail flange and all marine applications.

Biolubri GreaseKote 100 is an Environmentally Acceptable Lubricant (EAL) Certified under SS 155470 Program Certified an Environmentally Acceptable Lubricant (EAL) under the Swedish SS 155470 Labelling Program.[9]

It is a biodegradable, non-toxic grease that offers the ultimate protection against corrosion, rust and long-life lubrication under shipping conditions anywhere in the world. Developed for harsh marine environments, it provides an excellent alternative to mineral-based lubricants without comprising on performance and capabilities. It is suitable for applications where environmental parameters are a priority.

In all these industries, you have a clear loss of lubrication situation where the lubricating grease eventually ends up either in soil or water. Today, most modern bio-greases are formulated with different biodegradable synthetic esters. These products may actually have increased levels of performance compared to their mineral oil-based counterparts giving the bio-grease a clear edge.

A really interesting opportunity for bio-greases has recently occurred due to new legislation. The 2013 Vessel General Permit (VGP) issued by the United States Environmental Protection Agency. The VGP aims to reduce the environmental impact of ships, barges, tugs and other commercial vessels, protecting American coasts, lakes and rivers. For equipment such as pitch propellers, thruster bearing and wire ropes, the VPG points out that “

Biolubri Greasekote 100 meets OECD 301 F Biodegradability Test and OECD 201 Toxicity Test. For grease formulations with at least 75% (w/w) of constituent

substances biodegraded by at least 60% within 28 days according to OECD 301F. 25% (w/w) of the formulation need not meet the above biodegradability requirement and may have constituent substances that are either inherently biodegradable or non-biodegradable but may not be bio-accumulative.

To minimize confusion in the marketplace and to increase public awareness and create sensitivity for environmentally preferable products, national and international labeling programs have been developed, primarily in Europe (Habereder et al., 2008). These labeling programs have defined and established methods to measure the properties of a lubricant that would qualify it as being environmentally acceptable. The labeling programs can aid the purchasing decisions of a vessel operator by helping to remove uncertainty. The principal national and international labeling certification programs for bio-lubricants and EALs are presented below.

NATIONAL LABELING PROGRAMS

Blue Angel

The first national labeling scheme for lubricants was the German Blue Angel label, developed in 1988. Criteria have been developed for several classes of lubricants, including hydraulic fluids, lubricating oils, and greases. In order to qualify for certification, a lubricant must possess the following characteristics: biodegradability; low toxicity to aquatic organisms; non-bio-accumulative; and no dangerous components (such as carcinogens or toxic substances as defined by Germany's Ordinance on Hazardous Substances).

A product must also pass technical performance characteristics appropriate for its use. Biodegradability can be demonstrated using OECD tests 301B-301F to measure ultimate biodegradability or CEC L-33-A-934 to measure primary biodegradability.

Blue Angel's requirement for ultimate biodegradability is the primary difference between the Blue Angel labeling certification program and other national and international certification programs. Aquatic toxicity is determined according to OECD 201-203.

Products receiving the Blue Angel certification must also pass a series of technical performance requirements that depend on the class of lubricant. Unlike some of the other labeling programs, the Blue Angel certification does not have any requirements for renewability; consequently, lubricants comprised completely of petroleum-sourced components can receive Blue Angel certification. Nevertheless, Blue Angel

certification is considered rather stringent, and the proportion of lubricants receiving this certification remains low, with the majority being hydraulic fluids (Habereder et al., 2008). A complete list of all lubricants that carry the Blue Angel certification can be found at http://www.blauer-engel.de/en/products_brands/ search_products /search_for_products.php.

In Europe classifications have evolved locally introducing such standards as the German Blue Angel or **the Swedish Standard SS 155470**. [5,6,7].Today we have the Eco-label, governed by the European Union, which ties all European Standards together. Lubricants have their own group with their own criteria and to qualify should demonstrate compliance in-line with the following elements-No R-phrases, Aquatic toxicity requirements.

Biodegradability and bio-accumulative potential, Exclusion of specific substances, Renewable raw materials and last but not least Technical performance. The technical performance for greases is defined as “Fit for purpose”. A bit vague but still. This certification system gives governmental agencies and legislators a really powerful reference when discussing or enforcing the use of bio-lubricants. At the same time, it gives bio-grease manufacturers the opportunity to register conforming products and use the Eco-label in their marketing. This opportunity has been available for some years now but unfortunately the list of approved products is still surprisingly short. The VGP opportunity

Swedish Standard 155470

Another national labeling scheme for lubricants is the Swedish Standard, which includes standards for hydraulic fluids (SS 155434) and greases (SS 155470). Evaluation of a lubricant under the Swedish Standard involves testing for biodegradability and aquatic toxicity, as well as sensitizing properties of a lubricant formulation and its components (Habereder et al., 2008). The Swedish Standard evaluates biodegradability using ISO test methods (e.g., ISO 9439), and has varying requirements, depending upon class, for renewable resources content (SP 2010).

The Biodegradability and Toxicity Tests Biolubri GreaseKote 100 were tested by a third party independent laboratory Hygiene Institut des Ruhrgebiets in Germany.

Test Procedure for OECD 301 F Biodegradability[10,11,12] and results in Test Report 1 are herein described in procedure below:

1.1 Test conditions

1.1 Method description

Based on the method stipulated in the regulation OECD 301 F, 250ml of the possibly diluted substance, 1 ml of mineral nutrient salt solution as well as 10ml of activated sludge are prepared inside the reactors and incubated for 28 days at 25° C ± 1° C. The carbon dioxide produced during the degradation of the organic substance is chemically bonded to potassium hydroxide and the consequently developing low pressure in the system is manometrically measured. The quantity of oxygen used during the aerobic conversion of the organic matrix is equivalent to generated carbon dioxide concentration.

A preparation of 250ml of de-ionized water, 10ml of activated sludge and 1ml of mineral salt solution serves as a so-called blank trial. A standardized sodium benzoate solution is subjected to the entire test procedure for verification purposes.

1.2 Test organisms

Polyvalent microorganisms from activated sludge of a sewage plant cleaning waste water of predominantly domestic origin are used as test organisms.

The inoculum used in the present case was activated sludge 03.09.2014, which after 3 hours of ventilation was used till on the day of sampling. Hence, the sludge flora was adapted. To each preparation 10ml of activated sludge were added corresponding to 30mg of activated sludge dry substance per litre of the test preparation.

1.3 Chemicals

1.3.1 Mineral nutrient salt solution

Solution 1

8.5 grs potassium dihydrogen phosphate (KH_2PO_4), 21.75 grs dipotassium hydrogen phosphate (KH_2PO_4), 33.4 grs disodium hydrogen phosphate dihydrate (Na_2HPO $2H_2O$) and 0.5% grs ammonium chloride are diluted with distilled water to a litre.

Solution 2

22.5 grs magnesium sulfate heptahydrate ($MgSO_4 . 7 H_2O$) are diluted with distilled water to a litre.

Solution 3

36.4 grs calcium chloride dihydrate ($CaCl_2 . 2 H_2O$) are diuted with distilled water to a litre.

Solution 4

0.25 grs iron (lll) chloride hexahydrate ($FeCl_3 . 6H_2O$) are diluted with distilled water to a litre.

For preparation of the reday-to-use mineral nutrient medium 10 ml of the solution as well 1 ml of each of the solutions 2 to 4 are to be replenished with distilled water to a litre.

1.3.2 Standardized sodium benzoate solution

100 mg sodium benzoate solution are replenished with distilled water to a litre (theoretical oxygen demand- ThOD corresponding to 167 mg/l)

Test Solutions

A) Product test preparation
10 ml of activated sludge and 1 ml of nutrient salt solution were added to 25 ml test solution consisting of the product "Emulsified Biolubri Greasekote 100" (COD= 1022000 mg/l) diluted 1:10000 (COD corresponding to 106 mg/l).

B) Blank value preparation
The blank value preparation contained 250 ml od de-ionized water, 10 ml of activated sludge and 1 ml of nutrient salt solution.

C) Check preparation
A 250 ml sodium benzoate solution (ThOD = 167 mg/l of activated sludge and 1 ml of nutrient salt solution added served as check preparation.

1.5 Other test conditions

1.5.1 pH-values of the solutions

The pH-values measured at the start and at termination of the tests are indicated in the table below

	pH Values	
	start of the test	**end of the test period (28 days)**
Preparation A	7.4	7.5
Preparation B	7.4	7.5
Preparation C	7.4	7.5

1.5.2 **Duration of the test**

The duration of the test in this case was 28 days

1.5.3 **Test temperature**

The test temperature was at 25° C ± 1° C

1.5.4 **Device conditions**

The respirator was kept closed during the entire testing method. The absorption of the carbon dioxide formed by the oxidation of the organic ingredients of the test solution and liberated into gas phase was effected by a 45 percent potassium hydroxide solution in the CO_2 absorber in the upper part of the vessel. The pressure drop in the test vessel was indicated by a manometer and recorded daily.

1.5.5 **Identification of by-products from the degradation of the product "Emulsified Biolubri Greassekote 100"**

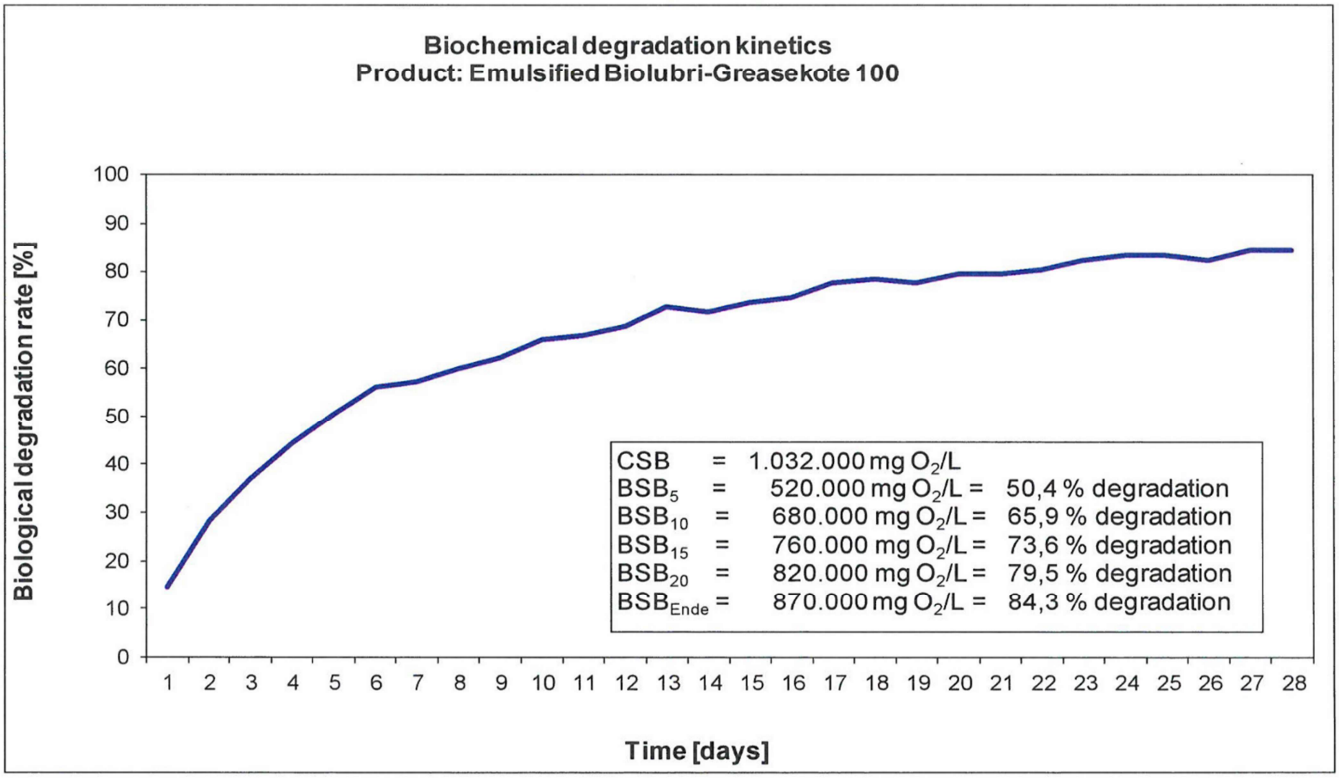
An identification of by-products originating from the degradation of the product "Emulsified Biolubri Greasekote 100 has not been carried out.

2. **Results and discussion**

The corresponding chemical oxygen demand (CDO) determined according to DIN 38409 or, respectively, the theoretical oxygen demand (ThOD) of the check preparation served as a value of reference and calculation with respect to biochemical degradation; the blank value has been accounted for by means of subtraction of the corresponding manometric readings.

Considering the chemical oxygen demand of 1032000 mg O_2 per litre determined for the product “Emulsified Biolubri Greasekote 100” as the quantity of oxygen required for the total degradation, the biochemical degradation after 5 days expressed as BOD is 520000 mg O_2 per litre = 50.4 per cent.

As it can be seen in the attached graphical representation of the manometrically determined biological degradation kinetics, the microbiological degradation of the biochemically oxidizable ingredients after 27-28 days reaches approx.84.3 percent



Test Procedure for OECD 201 Aquatic Toxicity and results are herein described in procedure below:

Sample of Biolubri Greaskote 100 was sent to Hyginene-Institut des Ruhrgebiets, based in Germany for Aquatic Toxicity Test using OECD 201 Test Method.

Test Procedure for OECD 201 [13,14,15] Aquatic Toxicity and Results are herein described in procedure below:

Minimally-toxic Formulation must pass OCED 201, 202, and 203 tests for acute toxicity testing. If a substance is evaluated for the formulation and main constituents, the LC50 of greases must be at least 1000mg/L. See Test Report 2 below

Biolubri Greaskote 100 is a wire rope conditioner and lubricant which is low soluble in water. So, the OECD 201 test was performed following the OECD Guidance Document on Aquatic Toxicity Testing of Difficult Substances and Mixture (OECD, 2000), where a "water-accommodated fraction" (WAF) is made by stirring the test substance for 72 h in water (here: 3000 mg Biolubri Greaskote 100 in 1 litre pure water) and subsequently removing the insoluble portions.

The corresponding examinations have been made in all cases with the above named WAF-concentration of 3000 mg per litre.

Examination results: determination of algae toxicity

Determination of the inhibition effect of cell reproduction in green algae took place according to the OECD directive 201. The green algae *Scenedesmus subspicatus* was cultivated under defined conditions (23°C ± 2°C; permanent lighting at 8000 Lux) in a culture medium for 72 hours with different concentrations of the test sample. The toxicity of the test product was determined at specific times (24 h, 48 h and 72 h) by determination of the cell counts.

The results of the cell reproduction inhibition test are listed below:

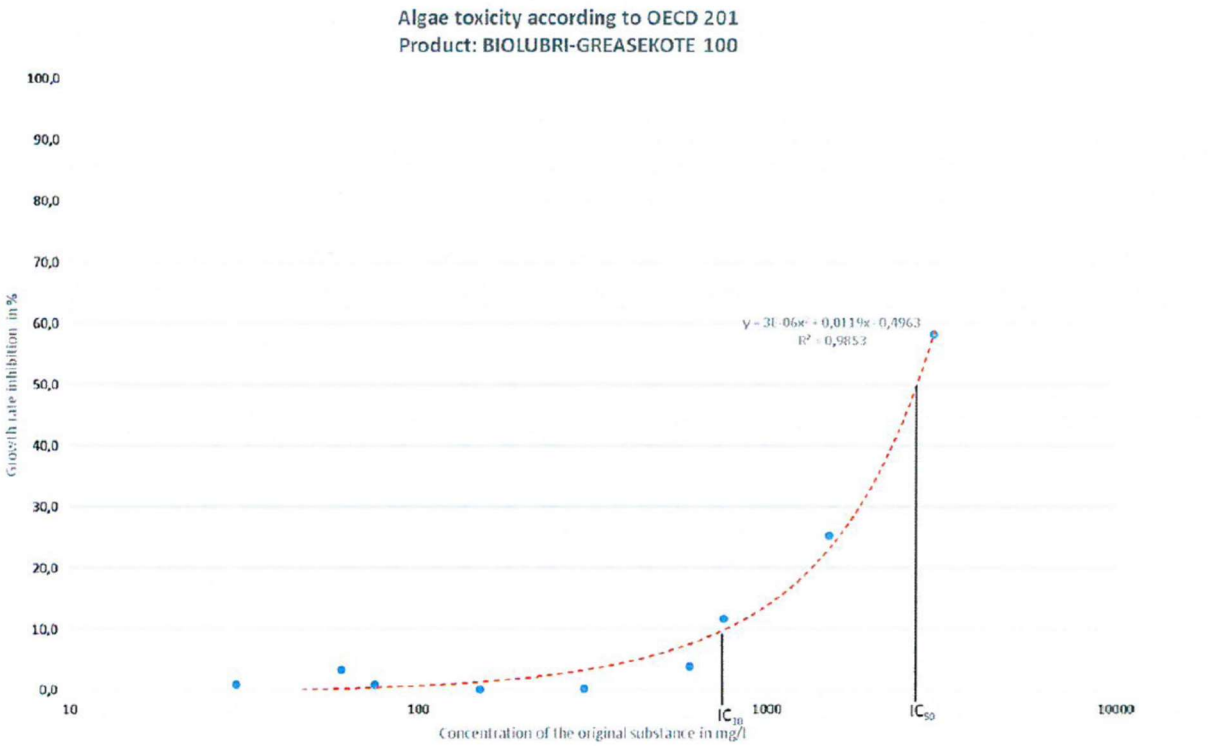
Concentration of the original substance in mg/l	30	60	75	150	300
Growth rate inhibition in %	<5	<5	<5	<5	<5

Concentration of the original substance in mg/l	600	750	1500	3000 (WAF)

Growth rate inhibition in %	<5	11.7	25.2	58.3

This shows the following effective concentrations (IC values) for the product "Biolubri Greaskote 100" in a graphical assessment (see below) at an inhibition of 10% and 50%.

IC10	(0 to 72 h)	=	750	mg/l
IC50	(0 to 72 h)	=	2700	mg/l



Test Report 2

Non-bio-accumulative Each non-biodegradable component of the formulation is tested to confirm its non-bio-accumulative properties.

Conclusion

Based on the above test results, Biolubri Greasekote 100 is considered an environmental acceptable lubricant (EAL) and is listed on the Swedish Standard SS155470 [1]

References

1) Lubricating greases which meet environmental requirements ...
https://www.sp.se/en/index/services/Lubricanting%20grease/Sidor/default.aspx
Lubricating greases which meet environmental requirements according to Swedish Standard SS 155470. The use of lubricating grease on moving parts is essential in order to save energy and diminish friction and wear. Since the grease used is consumed and may leak to the environment it is vital to use environmentally

2) 013 VGP Requirements for EALs
https://www.eagle.org/meagle/ShowProperty/.../References/.../ABSTrends_May2014 May 23, 2014 -according to the 2013 Vessel General Permit (VGP), effective 19 December 2013, all vessels (not only new vessels) must use environmentally acceptable lubricants (EALs) in all oil-to-sea interfaces, unless technically not feasible. All vessels were required to be in compliance by the effective date. The VGP.

3) Environmentally Acceptable Lubricants - epa nepis
https://nepis.epa.gov/Exe/ZyPURL.cgi?Dockey=P100DCJI.TXT
United States Environmental Protection Agency Office of Wastewater Management Washington, DC 20460 Environmentally Acceptable Lubricants EPA 800-R-11-002 ... Eco-label 17 5.2.3 OSPAR 19 5.3 Summary of Environmentally Acceptable Lubricant Labeling Programs 19 6 CONCLUSION 21 7 REFERENCES 7-22 11

4) 2013 Vessel General Permit (VGP) Factsheet - EPA
https://www3.epa.gov/npdes/pubs/vgp_fact_sheet2013.pdf
Dec 4, 2009 –Final 2013 VGP Fact Sheet. U.S. Environmental Protection Agency.2013 Final Issuance of. National Pollutant Discharge Elimination System. (NPDES) Vessel General Permit (VGP) for. Discharges Incidental to the Normal Operation of. Vessels. Fact Sheet. Agency: Environmental Protection Agency (EPA)

5) Blue Angel | Ecolabel Index
www.ecolabelindex.com/ecolabel/blue-angel

May 26, 2016 -The Blue Angel was initiated by the German government and awarded by an independent Jury to products that are environmentally friendlier than others serving the same use. Each label specifies that the product or service focuses on one of four different protection goals: health, climate, water, and..

6) From the Blue Angel to the EU Ecolabel - A meaningful environmental ...
https://www.eplf.com/.../blue-angel-eu-ecolabel-meaningful-environmental-label-euro... The Blue Angel ("Der blaue Engel") has made a name for itself among consumers as a quality label for low-emission products in many product categories, particularly in the German-speaking area, and has become a bra

7) Environmentally Acceptable Lubricants - Defense Technical ...
www.dtic.mil /dtic /tr /fulltext /u2 / a553091. pdf search products / search_for_products. php . 5.1.2 Swedish Standard. Another national labeling scheme for lubricants is the Swedish Standard, which includes standards for hydraulic fluids (SS155434) and greases (SS 155470). Evaluation of a lubricant under the Swedish Standard involves testing for biodegradability and..

8) Environmentally Acceptable Lubricants: Current Status and Future ...
www2.eng.cam.ac.uk/~mpfs/papers/articles/WTC2005/pdfs/t.../WTC2005-63503.pdf by NS Battersby - Cited by 18- Related articles World Tribology Congress III. September 12-16 2005, Washington D.C.. WTC2005-63503. ENVIRONMENTALLY ACCEPTABLE LUBRICANTS: CURRENT STATUS AND FUTURE OPPORTUNITIES. Nigel S. Battersby. Shell Global Solutions (UK), Cheshire Innovation Park, PO Box 1 Chester, United Kingdom. ABSTRACT

9) Lubricating greases which meet environmental requirements ...
https://www.sp.se/en/index/services/Lubricanting%20grease/Sidor/default.aspx
Lubricating greases which meet environmental requirements according to Swedish Standard SS 155470. The use of lubricating grease on moving parts is essential in order to save energy and diminish friction and wear. Since the grease used is consumed and may leak to the environment it is vital to use environmentally

10) OECD guideline for testing of chemicals - OECD.org
https://www.oecd.org/chemicalsafety/risk-assessment/1948209.pdf
by R Biodegradability - Cited by 5- Related articles
Jul 17, 1992 -biodegradability in an aerobic aqueous medium. They are: 301A: DOC Die-Away.301B: CO2 Evolution (Modified Sturm Test).301 C: MITI (I)

(Ministry of International Trade and. Industry, Japan).301D: Closed Bottle.301 E: Modified OECD Screening.301 F: Manometric Respirometry. Method 301A is...

11) OECD 301F – Biodegradation Test – O2 Consumption | Situ ...
www.situbiosciences.com/biodegradation/oecd-301f-biodegradation-test-o2-consumpt.OECD 301F is a solutions biodegradation test that determines the biodegradability of a material by measuring oxygen consumption

12) OECD iLibrary | Test No. 301: Ready Biodegradability
https://www.oecd-ilibrary.org/.../test-no-301-ready-biodegradability_9789264070349-e..This Test nuideline describes six methods that permit the screening of chemicals for ready biodegradability in an aerobic aqueous medium. The methods are: the DOC Die-Away, the CO2 Evolution (Modified Sturm Test), the MITI (I) (Ministry of International Trade and Industry, Japan ..

13) OECD Medium, Stock Solutions | Wako Information
www.wako-chem.co.jp › Home › Products › Analysis and Environment Jump to For preparation of 1 L of OECD Culture Medium based on OECD Test Guidelines 201, take the OECD Medium, Stock solution I ~ IV as shown below, mix together and dilute with ion-exchanged water, etc. to make the 1 L solution

14) OECD iLibrary | Test No. 201: Freshwater Alga and Cyanobacteria ...
https://www.oecd-ilibrary.org/.../test-no-201-alga-growth-inhibition-test_978926406992.The purpose of this test is to determine the effects of a substance on the growth of freshwater microalgae and/or cyanobacteria. Exponentially growing test organisms are exposed to the test substance in batch cultures over a period of normally 72 hours. The system response is the reduction of growth in a series of algal.

15) OECD guidelines for the testing of chemicals - OECD.org
www.oecd.org/chemicalsafety/testing/1946914.pdf O**ECD** Guidelines for **Testing** of Chemicals are periodically reviewed in the light of scientific progress. With respect to Guideline **201**, Alga, Growth Inhibition **Test** (adopted June 1984), the need to extend the Guideline to include additional species and update it to meet the requirements for hazard assessment.