Concept Paper

Reef Protection Factor: A Gold Standard for Coral-Safe Products

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Reef Protection Factor: A Gold Standard for Coral-Safe Products

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Abstract: Background: In recent years, the cosmetics industry (particularly the sunscreen sector), has made claims of "reef safe" products without validation, as no standardised verification methods or regulatory oversight currently exist. This has led to false and/or unsubstantiated marketing, which may have resulted in increased or unknown risks to the environment as well as greenwashing products to consumers. Indeed, 48% of products labelled as "reef safe" still contain known coral-toxic compounds. **Objectives:** Here, we present the Reef Protection Factor (RPF). A standardised three-tiered certification framework with bronze, silver, and gold ratings. RPF allows for the scientific evaluation and designation of a safety level that can be applied to any given cosmetic product indicating the impact it will most likely have on corals and other reef associated invertebrates. **Discussion:** The RPF evaluation protocol and ranking are transparent and rigorous. The method is practical to implement in the majority of facilities as the testing cost is low. We are hopeful that this will encourage widespread use of the RPF ranking system and ensure that the framework does not favour large multinational conglomerates over smaller, single-product or range-limited companies.

Keywords: reef protection factor; certification framework; reef safe; cosmetics

Introduction

Although many cosmetic products contain harmful ingredients that impact our natural world, it was the relationship between sunscreen exposure and coral health degradation that changed many people's way of thinking [1]. Indeed, after this first publication (2008) [1], there was a surge in alternative sunscreen formulations brought to market and labelled as "reef safe". However, the absence of regulated standards for such claims has resulted in potentially harmful ingredients still reaching sensitive marine environments, often without consumer knowledge [2]. 48% of sunscreens with the "reef safe" label still contain oxybenzone, octinoxate or other ingredients which have all been shown to be "reef toxic" by scientists and governing bodies [3].

Many of these "reef toxic" ingredients remain in our ocean's waters, especially in tourist-heavy reef areas - the origins via bathers, or indirectly through the likes of wastewater effluent after showering [4]. Oxybenzone contamination for example, has been shown to occur at concentrations from 0.8 μg/L to 1.4 mg/L [5]. Levels that are up to 140 times higher than concentrations known to negatively impact coral health [6]. This has now prompted some countries to implement product bans [7–11].

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Yet, banning products such as sunscreens is certainly not a straightforward process. Especially as this could be detrimental to human health. Therefore, products need to be developed that not only protect human skin from harmful UV radiation but are truly safe for our environment. Many have called for a standardised protocol for sunscreen testing [2,12,13], although there are a number of scientific publications which document the impact of sunscreen and other cosmetic products on corals, comparisons between studies and therefore between products remains impossible or difficult at best. They often vary in loading rates assessed, and exposure - across µg/L to mg/L, and hours to weeks [13]. There is also often variation in the species assessed, life cycle stages utilised, and number of genetically unique individuals tested, not to mention the response metrics measured [1,5,6,6,12,14–24].

Regulatory requirements are certainly not a novel idea and indeed exist for cosmetic safety assessments and their impact on humans. Take the cosmetics regulation N°1223/2009 as an example [25]. Across Europe, specific ingredients in cosmetics are evaluated by the Scientific Committee on Consumer Safety (SCCS), which publishes its evidence-based opinion on safe conditions of use. This is often alongside the General Product Safety Regulation (GPSA) managed by the EU Scientific Committee on Cosmetology. In the USA, the trade association with the support of the Food and Drug Administration (FDA), established the Cosmetic Ingredient Review (CIR), which prioritises and assesses cosmetic ingredients. However, CIR generally attributes risk to groups of similar substances based on chemical families or plant-derived ingredients and does not necessarily test each one. Interestingly, more evidence-based opinions and recommendations (such as those provided by the SCCS) are often heralded as "better practice" compared to providing prescriptive demands for strict adherence to precise regulatory "guidelines". Although, the SCSS appears to have a favourable opinion of many UV filters such as Octocrylene, primarily because of their known benefits to humans, they do not take into account the evidence that such compounds are detrimental to our environment and trigger disorders such as cell mitochondrial dysfunction in a wide range of organisms [21]. Thus, these regulatory processes urgently need updating to not only account for the safety of humans, but for the safety of other organisms where the cosmetic in question may have an impact.

Although we focus on sunscreens, as this is where the majority of "reef safe" products are emerging, other cosmetics such as shampoos, soaps, and laundry detergents will undoubtedly impact reef environments around the world. Indeed, ingredients such as Benzophenone-2 and Oxybenzone (products in many different cosmetics) have been shown to negatively impact the health of reef organisms [5,16]. So, with this in mind any such regulatory framework proposed should, where possible, be universal in its application across a range of products.

Discussion

Here, we present the Reef Protection Factor (RPF): a standardised, three-tiered certification framework with bronze, silver, and gold ratings. RPF allows for the scientific evaluation and designation of a safety level that can be applied to any given cosmetic product, indicating the impact it will have on corals and other reef-associated invertebrates. This accreditation factor not only considers rigorous scientific testing but is practical for multiple laboratories to undertake due to its cost effectiveness.

In general, we recommend the use of multiple species and genetically distinct individuals for testing all stages. The water-accommodated fraction (or WAF) is the recommended method for loading the cosmetics into the tanks. However, in some cases, use of an artificial skin may be necessary. Acute tests (giving a bronze accreditation) run for seven days, whilst the chronic tests for silver and gold run for 50 and 100 days respectively. If any signs of ill health are witnessed in any of the test subjects, the cosmetic does not qualify (this includes, mortality, tissue loss, colour change, feeding rate disruption, polyp retraction etc). A more detailed description of the specific requirements and steps necessary to be awarded any accreditation can be found below.

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Standardisation of Environmental Parameters

To be eligible for certification of any rating, the experimental systems used for the test must meet the criteria outlined in Table 1. This ensures that water quality remains suitable and stable for the organisms and does not confound the results.

Table 1. Environmental parameters with target ranges and minimum testing frequency.

Validation Criteria	Target Range	Minimum Testing Frequency
Temperature	25-27°C	Daily *
Salinity	33–38 ppt	Daily *
Light	100–500 μEm^{-2} s^{-1}	Daily *
pH	7.8-8.4	Weekly **
Alkalinity (or the related carbonate hardness, KH)	7-9 dKH	Weekly **
Calcium	380-450 ppm	Weekly **
Magnesium	1200-1400 mEq/L	Weekly **
Phosphate	0.02-0.1 ppm	Weekly **
Ammonia	≤0.1 ppm	Weekly **
Nitrates	2-10 ppm	Weekly **
O_2 Saturation	>75%	Weekly **
Day night cycles (hours)	12: 12	
Water change	10-15% per week ***	
Food	Three times per week. Suggested foods include live zooplankton, brine shrimp nauplii (*Artemia salina*), or commercially available coral feed. Other marine invertebrates must also be fed suiting their dietary requirements.	

* Minimum of five times/days per week during chronic (>7 days) tests. ** During acute (7 day) tests these must be taken at the start and end of the experiment as a minimum instead of weekly. *** Does not apply to acute (7 day) tests.

Life Stage of Coral and Species Considerations

Although there has been some effort to include coral larvae when testing UV filters [5,6,16], we propose that the use of adult corals is more suitable for RPF as they are more widely available, relatively easy to culture, and easily replicable via asexual fragmentation. Further, urchins and snails would make good test subjects, due to their ease of culture and upkeep and use in previous sensitivity trials [26,27]. The RPF certification protocol has been optimised to be suitable for: 1) multiple species representing several families, and 2) multiple genetically unique coral fragments or invertebrate individuals per species per tank. Incorporating multiple organisms of each species captures the natural variation in responses to various stressors for any given species or even genotype. When fragmenting parental colonies of corals, a minimum of 30 days should follow to ensure no disease or tissue loss occurs. [28]. Further, an acclimatisation period of at least seven days should be undertaken for all invertebrates in the test aquaria before any dosing is undertaken [28].

Application of Product

Currently, water-accommodated fraction (WAF) is the only known method for testing the toxicity of an entire complex substance in a liquid [29]. and has been adopted for the application of cosmetic products for testing when applying for bronze, silver and gold accreditation. Loading rates

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refer to the total amount of the test substance added to the test medium to prepare the WAF [30]. Here we recommend a loading rate based on the expected environmental concentration of sunscreen, 30 mg/L, released by 100 beachgoers [31]. The cosmetic of interest should be dissolved into the treatment seawater to a concentration of 30 mg/L in a lidded glass vessel and stirred for 24 hours [30]. Should the product be at risk of photodegradation they should be protected from light during this period [30]. Other cosmetics such as shampoos, conditioners would likely be released in varying quantities per beach goer. Thus, the acute test loading rate may need to be modified according to the concentration of toxic components (or active ingredients) present in the cosmetic under review. For hydrophobic mixtures, up to 48 hours may be needed to fully dissolve the product, however product stability and risk sorption to the vessel should be considered [30]. This cosmetic solution is then transferred into the treatment tanks. In line with the ISO 24444:2019 SPF testing guidelines, there is a maximum acceptable error of ± 2.5 % in the loading rate of the tested product. An identically treated cosmetics-free batch of seawater must be added to the control tanks at the same time.

In some cases, the cosmetics due to be tested are designed to be insoluble, remaining on the skin for longer periods, and needing less re-application. However, this presents challenges for standardisation as the WAF method is likely to be ineffective or at best incomparable to other more soluble solutions. In these cases, artificial skins can be used for the application of the products. Various types of artificial skin have been developed for use in Sun Protection Factor (SPF) testing, some of which have similar properties to human skin [32,33]. We recommend that the sunscreen-covered artificial skin should be left in the treatment tanks for 24 hours and this step repeated every three times per week. The loading rate using artificial skins needs to be adjusted compared to the WAF method as least 25% of sunscreen applied to the skin is expected to wash off during swimming or bathing [1]. Therefore we recommend the artificial skin be dosed at 120 mg/L ± 2.5 % total product. The product should be spread across the artificial skin at a thickness no lower than 2 mg/cm^2, which is the FDA recommending amount of sunscreen to be applied per person [1]. An identical artificial skin (without sunscreen or other cosmetic product) must be placed into the control tanks at the same time as the treatment tanks.

Inductively Coupled Plasma Optical Emission Spectroscopy (ICP-OES), Inductively Coupled Plasma Mass Spectrometry (ICP-MS), High Performance Liquid Chromatography with Ultraviolet (HPLC-UV), Liquid Chromatography Tandem Mass Spectrometry (LC-MS/MS), High Performance Liquid Chromatography, Gas Chromatography or Atomic Absorption Spectrometry (AAS) can be used to quantify the amount of the cosmetic's active ingredient in the tank seawater during testing [30,34–37]. Here, the choice of spectrometer will depend on the type of cosmetic ingredients being quantified. Ultraviolet Absorption Spectrophotometry (UAS) may also be used to quantify total sunscreen concentrations [31].

Bronze Accreditation

The entry-level certification requires a seven-day acute toxicity assessment (Table 2). The invertebrates are exposed to the cosmetic thrice weekly either via the WAF method at 30 mg/L, or the artificial skin method at 120 mg/L. A minimum of three independently running tanks per treatment (and per control) are needed to account for potential tank effects, and each tank must meet the validation criteria in Table 1 for the duration of testing. While the acute tests minimum tank size requirement is 3.8 L, larger tanks are recommended to maintain stability in the water parameters and stocking densities based on the size and number of the species/fragments undergoing treatment [20]. The minimum volume of 3.8 L reduces the risk of dramatic swings in water chemistry that are known to occur with smaller volumes. Artificial aeration or shaker tables may help maintain oxygen levels and enhance diffusion required to meet the validation criteria (Table 1). At a minimum, the tank seawater concentrations of the cosmetic being tested should be measured immediately prior to the first dose, within one hour of the first dose, 24 hours after the first dose and seven days after the first dose. In addition, seawater chemistry must be measured at the start and end of the testing period.

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The effect of cosmetic exposure on corals will be assessed by measuring polyp retraction, tissue loss, mucus hypersecretion, paling, and mortality. The effect of cosmetic exposure on other invertebrates will be measured using overall health state (visual inspection, including colour, food ingestion rate and mortality). If any of these visual signs of distress are observed on any of the test subjects, the product does not pass this accreditation and is not classified as "reef safe".

Table 2. Summary of the test requirements for each RPF accreditation level.

	RPF Reef Protection Factor	RPF Reef Protection Factor	RPF Reef Protection Factor
Accreditation	Acute toxicity assessment with none of the following occurring during the texting period: 1. Mortality 2. Tissue necrosis 3. Polyp retraction 4. Tissue discolouration 5. Mucus hypersecretion If any of the tested organisms show any of the acute responses in the control group, the test will not be valid.	Chronic toxicity assessment with none of the following occurring during the testing period: 1. Mortality 2. Tissue necrosis 3. Polyp retraction 4. Tissue discolouration 5. Mucus hypersecretion 6. Significant reduction in growth rate 7. Significant reduction in endosymbiotic algal density or PSII yield. If any of the tested organisms show any of the acute responses in the control group, the test will not be valid.	Chronic toxicity assessment with none of the following occurring during the testing period: 1. Mortality 2. Tissue necrosis 3. Polyp retraction 4. Tissue discolouration 5. Mucus hypersecretion 6. Significant reduction in growth rate 7. Significant reduction in endosymbiotic algal density or PSII yield If any of the tested organisms show any of the acute responses in the control group, the test will not be valid. The product tested must have an additive benefit to a species health (such as an increase in growth) to gain gold accreditation.
Test Duration (days)	7	50	100

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Dosing Concentration	On days 1, 3, and 5 add 30 mg/L dose using WAF method OR 24 hours of 120 mg/L on artificial skin.	On days 1, 3, and 5 of each week add 30 mg/L dose using WAF method OR 24 hours of 120 mg/L on artificial skin.
Dosing Method	WAF or artificial skin	
Tanks	- 3 x 3.8L or larger independent closed system tanks per treatment. - Starting invertebrate biomass: seawater < 1 g wet weight soft tissue/L [30].	- 3 x 50L or larger independent closed system glass tanks per treatment tank. A lower volume can only be achieved with suitable header tanks. Adequate water movement should be provided in each tank. - Starting invertebrate biomass: seawater < 1 g wet weight soft tissue/L [30].
Species	- Five species from five different genera of which three or more species are Scleractinian corals (at least one branching and one non-branching morphology). - Four genotypes per species must be used in each treatment.	
Feeding	- Scleractinian corals should be fed at least three times a week with one of live zooplankton, brine shrimp nauplii (*Artemia salina*), or commercially available coral feed. - Other marine invertebrates fed suiting their dietary requirements.	
Fragmentation and Acclimatisation	- Fragmentation 30 days prior to the start of testing - Corals acclimated to the testing vessels seawater for 7 days prior to testing.	
Health Monitoring	Tissue necrosis, polyp retraction, tissue paling at the start and end of the testing periods. Coral watch health charts can be utilised, or assessment of photographs using grey-scale normalised intensity values.	
Growth Monitoring	Not applicable to bronze accreditation.	Difference in linear and horizontal growth measured in 2D or 3D photogrammetry, and/or wet weight.
Symbiosis stability	Photographs, endosymbiotic algal cell counts, and/or pulse amplitude modulated (PAM) fluorometry.	
Repeat experiments	The test subjects used to gain RPF accreditation should not be "reused" for further testing if a normal health state is uncertain or within one month (30 days). Bronze accreditation is always recommended before applying for the higher levels.	

Silver Accreditation

Silver accreditation has the same experimental setup as bronze but runs for a minimum 50-day chronic exposure (Table 2), aimed at evaluating longer-term effects on invertebrate growth and health status. Same as for bronze, the test subjects are exposed to the product thrice weekly (either via the WAF method at 30 mg/L or the artificial skin method at 120 mg/L as described above). This RPF accreditation includes increased monitoring and reporting. At a minimum, the tank seawater concentrations of the cosmetic being tested should be measured immediately prior to the first dose, within one hour of the first dose, 24 hours after the first dose and 50 days after the first dose. In addition, weekly seawater chemistry levels are required. Due to the longevity of this study, tank size

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is larger (minimum 50 L), and the water needs to be changed by 10-15% per week. Same as for bronze, a minimum of three treatment and three control tanks is needed. In cases where header tanks are utilised, smaller treatment tank volumes of 5-10 L can be utilised - however again space for the required number of corals (and other invertebrates) needs to be considered [23].

The longer-term effects of cosmetic exposure on corals will be assessed by measuring growth and algal symbiont density or photosynthetic efficiency of PSII. These are in addition to the health metrics for bronze accreditation (polyp retraction, tissue loss, mucus hypersecretion, paling, and mortality). The effect of cosmetic exposure on other invertebrates will be measured in a similar manner. Growth of the corals can be determined by measuring the change in linear and horizontal dimensions acquired from 2D photos or 3D photogrammetry and/or from changes in wet mass between the initial and final time points of the experiment [38,39]. To assess changes in the coral-algal symbiosis, (known to be disrupted by pollution, light and heat for example)[40,41]. we can measure algal endosymbiont density via comparing and contrasting photos across the tanks[42]. and/or by direct microscopic counts on coral sub-samples at the end of the study [43]. To quantify photochemical efficiency, pulse amplitude modulated (PAM) fluorometry can be used to measure PSII yields [44]. Importantly, coral-algae relationships are complex and so these measures should not be taken in isolation. An increase or decrease in algal density or PSII yields can mean a disruption in health [45,46], so care needs to be taken when interpreting the results for these measures.

A cosmetic can be awarded Silver accreditation if the test subjects show no statistically significant negative difference in key health measures such as growth when treatments are compared against controls.

Gold Accreditation

The highest accreditation level represents the gold standard in “reef safe” product validation. Gold accreditation has all the same requirements as the silver testing (up until day 50) and must also see no negative effect in any metric measured. It then extends testing to 100 days (Table 2). On day 100±1 there must be an additional measure of the concentration of the product in the tank water, along with the test subject health metrics.

To warrant Gold accreditation, there must be an observable (and statistically significant) additive benefit to key health metrics such as growth for at least one of the species tested.

Implementation and Cost Considerations

Assuming those seeking certification already have a fully equipped laboratory and seawater tank system, and the cosmetic in question would be provided by the manufacturing company, we estimate the RPF certification rating costs range from approximately £3500 to £12,000. Cost estimates include consumables such as salt, buffers, invertebrates, and water tests as well as labour. Predictions for these costs are displayed in Table 3.

Table 3. Predictions of basic costs of each accreditation rating in 200 L tanks.

			
Corals/Invertebrates *	£2000	£2000	£2000

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Salt	£250	£450	£600
Concentration measurements (e.g., ICP-OES)	£558	£750	£1000
Labour (at minimum wage in the UK) *	£500	£4000	£8000
Buffers (e.g., Kh, Calcium and Magnesium) *	£50	£50	£50
Water tests *	£80	£80	£150

* Costs may be reduced by using material across multiple tests.

Additional costs of site rental, electricity, and water must be considered, as well as general equipment costs needed for each test such as a 3D scanner and modelling license, a PAM fluorometry machine, and tanks. As such the cost of the RPF certification will vary depending on the laboratory running the accreditation tests.

Wastewater from RPF testing must be disposed of responsibly and in accordance with local regulations. To ensure transparency and consumer trust, the reports of all successful tests must be made publicly available when certified by a newly established independent board. We have produced a landing page for this accreditation (https://rpf.world/) and it should be noted RPF is not for profit, although the accredited testing laboratories may wish to charge for the tests undertaken appropriately. Importantly, RPF certification applies only to the specific tested product and in the case of sunscreens the SPF levels. Formulations with lower or different SPF, or other products within the same product line, require separate testing and independent certification.

Conclusion

This standardised protocol addresses the historical inconsistencies in cosmetic-coral toxicity testing, while simultaneously maintaining practical feasibility for manufacturers. The tiered accreditation system provides flexibility for different levels of commitment to reef safety while ensuring meaningful environmental protection standards. By establishing clear, quantifiable criteria for "reef safe" claims, this protocol aims to protect coral reef ecosystems while providing consumers with reliable product information.

The RPF protocol represents a significant step towards standardising the evaluation of genuinely "reef safe" cosmetic products. Through its tiered approach and comprehensive testing requirements, it provides a practical framework for manufacturers while ensuring robust environmental protection standards. The successful implementation of this protocol could significantly reduce the impact of cosmetic products on coral reef ecosystems while restoring meaning to "reef safe" product claims.

Conflicts of Interest: Julia Rücker and Christian Wild are affiliated with the University of Bremen and Triona Barker and Michael Sweet are affiliated with the University of Derby, these universities will be involved in offering services for testing cosmetics for the RPF accreditation going forward. Michael Sweet is a co-founder of Ocean Guard, which will be the company providing not-for-profit RPF accreditation based on reports from the testing laboratories.

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