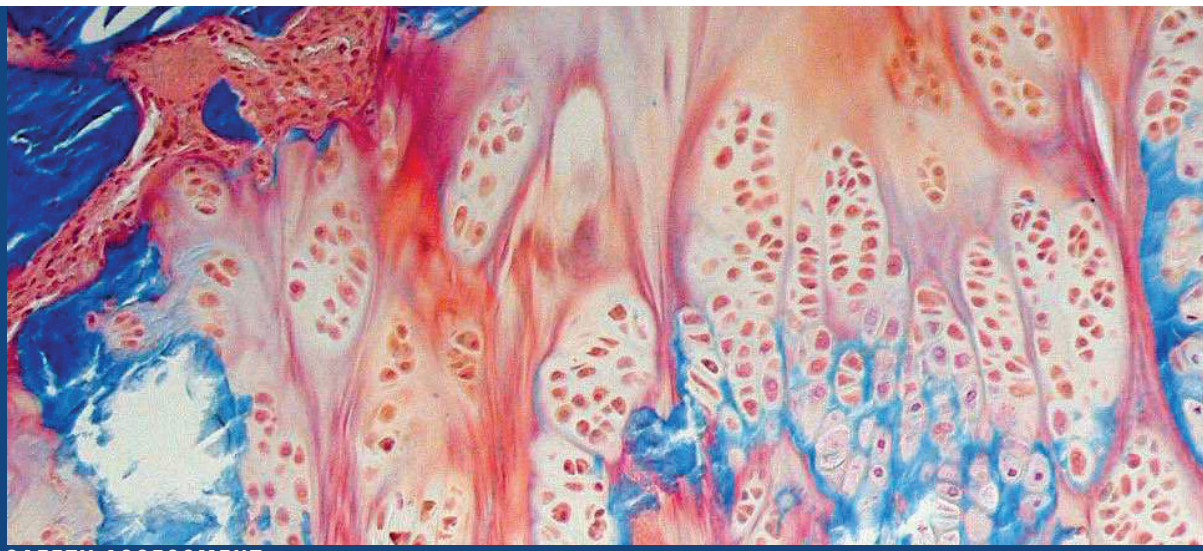
charles river

Summary

Charles River offers a multitude of *in silico*, *in chemico* and *in vitro* models to assess toxicity of drugs and chemicals as a screen or as a replacement to well established animal toxicology tests.



SAFETY ASSESSMENT

In Vitro Toxicology Models

Charles River is firmly committed to the 3Rs (Replacement, Reduction and Refinement) as the founder company of the North American 3Rs Collaborative (www.NA3RsC.org). The pursuit of this mission includes our development and adoption of replacements to animal testing, whenever possible. Many acute toxicity tests have been, or are being, effectively replaced by *in vitro* human, animal and 3D tissue alternatives. This shift is particularly evident in dermal irritation, corrosion, sensitization and phototoxicology, skin absorption and ocular irritation and severe damage. In addition, we are developing models to test toxicities and efficacy in other tissues such as the lung, mouth, and gut.

Ocular Irritation and Severe Damage

There is no single *in vitro* test available to replace the *in vivo* Draize test (OECD 405). The bovine corneal opacity and permeability (BCOP) test (OECD 437) and EpiOcular™ or HCE 3D models (OECD 492) can be used together to classify many Class 1 chemicals and chemicals not requiring classification for eye irritation or serious eye damage. Where a classification is required and the *in vitro* models do not give a clear overall prediction, the results of these tests can be used to justify performing the *in vivo* test.

Skin Corrosion

Skin corrosion can be assessed in a top-down or bottom-up testing strategy using the *in chemico* membrane barrier test method for skin corrosion, Corrositex™ (OECD 435), and the *in vitro* EpiSkin® or EpiDerm™ skin corrosion assay (OECD 431).

Skin Irritation

Changing the study design (OECD 439), but still using EpiSkin® or EpiDerm™, can additionally classify substances as irritants in accordance with GHS Category 2 or as No Category.

Skin Sensitization

A battery of tests are utilized as either screens or replacements for the *in vivo* skin sensitization assays (i.e., LLNA and Buehler tests). Initially, the *in silico* Lhasa Derek Nexus software is used. The *in chemico* DPRA (OECD 442C), *in vitro* KeratinoSens™ or LuSens (OECD 442D) and U SENS™ (OECD 442E) tests are performed in series. A prediction model is then used to classify the chemical as sensitizer or non-sensitizer. Currently, there is no non-animal test that fully provides a potency prediction. Therefore, weight of evidence and non-validated test models, for example GARDskin™, can be used. Finally, these screens may be used to justify the *in vivo* tests for potency.

SENZA GEN

Phototoxicity

The ultraviolet-visible radiation absorption spectrum test (OECD 101) can be used to assess whether there is a trigger for photosafety testing. The 3T3-NRU assay (OECD 432) is an *in vitro* cellular assay to determine phototoxic potential of a test article or chemical and, when the result is negative, is recognized as predictive of an *in vivo* negative response, serving as a direct replacement for *in vivo* preclinical phototoxicity assays. *In vitro* 3D keratinocyte systems can also be used to evaluate phototoxic potential of topical formulations that cannot be evaluated using the 3T3 assay. When required, an *in vivo* test can be performed. The melanin binding assay may be used to choose whether a pigmented or non-pigmented animal should be used.

Skin Absorption

Skin absorption testing (OECD 428) is performed in human, toxicology, and large animal species to assess the amount of chemical or drug that penetrates through or is located in the skin. Where required, the rat *in vivo* assay (OECD 427) may be performed usually to identify the fate of the stratum corneum reservoir bound material. These tests are used in safety testing of agrochemicals, chemicals, biocides, and drugs, and also in lead selection of topical formulations and in bioequivalence testing of biosimilars.

Pulmonary Toxicology

The EpiAirway™ and MucilAir™ models for the upper airway tract can be used to provide information on toxicity, or in support of dose range finding for animal models as a component of 3Rs and integrated toxicology testing. Diseased human as well as rat-derived models are also available for investigational and translational toxicity testing.

Cytotoxicity

The cytotoxicity assay in 3T3 cells (OECD 129) is used to estimate starting doses for acute oral systemic *in vivo* toxicity tests. Cytotoxicity tests are also required for medical devices (ISO 10993, part 5).

Development of New Assays and Investigational & Mechanistic Toxicology

Charles River is a member of EU-NETVAL and has a long history of evaluating, transferring, and establishing fit-for-purpose validations of assays. This knowledge has been used to generate bespoke or one-off assays for individual client needs. This has been developed into an investigational and mechanistic toxicology service utilizing 1D, 2D, 3D, and tissue models. These models can predict toxicity and increasingly, efficacy and, where possible, aid translational toxicology.

Scientists

Charles River scientists are always prepared to advise on the most suitable tests and testing strategy. They are supported by the Regulatory Affairs and Scientific Advisory Service groups where additional guidance is also available.