

## **CAM** assay

The chorioallantoic membrane (CAM) is a highly vascularized extra-embryonic membrane that mediates exchange of gas and nutrients during embryonic chicken development. The CAM model is often used for vascular research but the CAM can also be used to graft tumors and to inject compounds into the circulation of the developing chicken. The latter can be used to test e.g., compound toxicity. The advantage of the model is that it is low in cost, reproducible, reliable and fairly simple to perform. In addition, the model does not require a sterile work environment and in the Netherlands no approval of the animal welfare committee is required.

With regard to radiotherapy, the egg/embryo can tolerate a single dose of max 5 Gy or daily 2 Gy fractions for up to 1 week. Combining irradiation with injection of drug compounds can be used to determine the possible toxicity effects of the combination by monitoring embryo survival during and after treatment up to embryonic day of development (EDD) 17.

An outline of the CAM assay is provided in the figure below.

**Tip**: The provided data can be used to generate survival curves which will give information on the toxicity of some compounds when used in combination with irradiation. Be aware that not all groups start with the same number of eggs so make sure to look at relative survival numbers.



## Overview of embryonic growth and experimental time window of the CAM assay.

Fertizlized eggs are incubated until EDD6. At that day, part of the egg shell is opened to expose the CAM. Treatment can also start on that day or on EDD10 when most of the vasculature has formed. For treatment, the entire egg can be irradiated and compounds can be carefully injected in one of the larger CAM vessels (see pictures on the right). Subsequently, the survival of the embryo can be daily monitored up to EDD17 at which the experiment is terminated.