Chromium release assay
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Principle:
• The assay measures the killing of target cells by specific CTL which recognize a class I - peptide combination on the surface of the target cells.

Objective:
• Measuring antigen presentation by the target cell
• Measuring the activity of a T cell clone

Procedure:
• Target cells are labeled by incubating them with radioactive sodium chromate (Na$_2$CrO$_4$). Cells take up the chromate ion and store it in the cytosol.
• T cells (= effector cells) are added to the target cells in different amounts (effector to target ratios) and incubated for a given time.
• If the T cells kill the target cells then the radioactivity is released into the supernatant of the cells.
• The tubes (or plate) are then centrifuged and an aliquot of the supernatant is analyzed for radioactivity in a gamma counter.
  ♣ If there is a lot of radioactivity in the supernatant that means many cells have been killed.
  ♣ If there is no radioactivity in the supernatant that means no cells have been killed.
• Controls:
  ♣ Positive control: Add detergent to the target cells. Counts as 100% killing.
  ♣ Negative control: Add no CTL. Counts as 0% specific lysis (background).
• The results are given as "% specific lysis".

![Diagram of Chromium release assay](Figure A-38 Immunobiology, 6/e, © Garland Science 2005)
Example:

The figure shows several chromium release assays.
In panel B, several concentrations of three different peptides are tested which, when added to cells, make them vulnerable to the specific CTL.
In panel D, dendritic cells (DCs) are added in increasing concentrations to CD8 cells (CTLs), and their lysis is observed.
In panel E, four different ways of sensitizing target cells for CTL lysis (two different peptides with and without added hsp70) are tested.