# Better ways to do research:

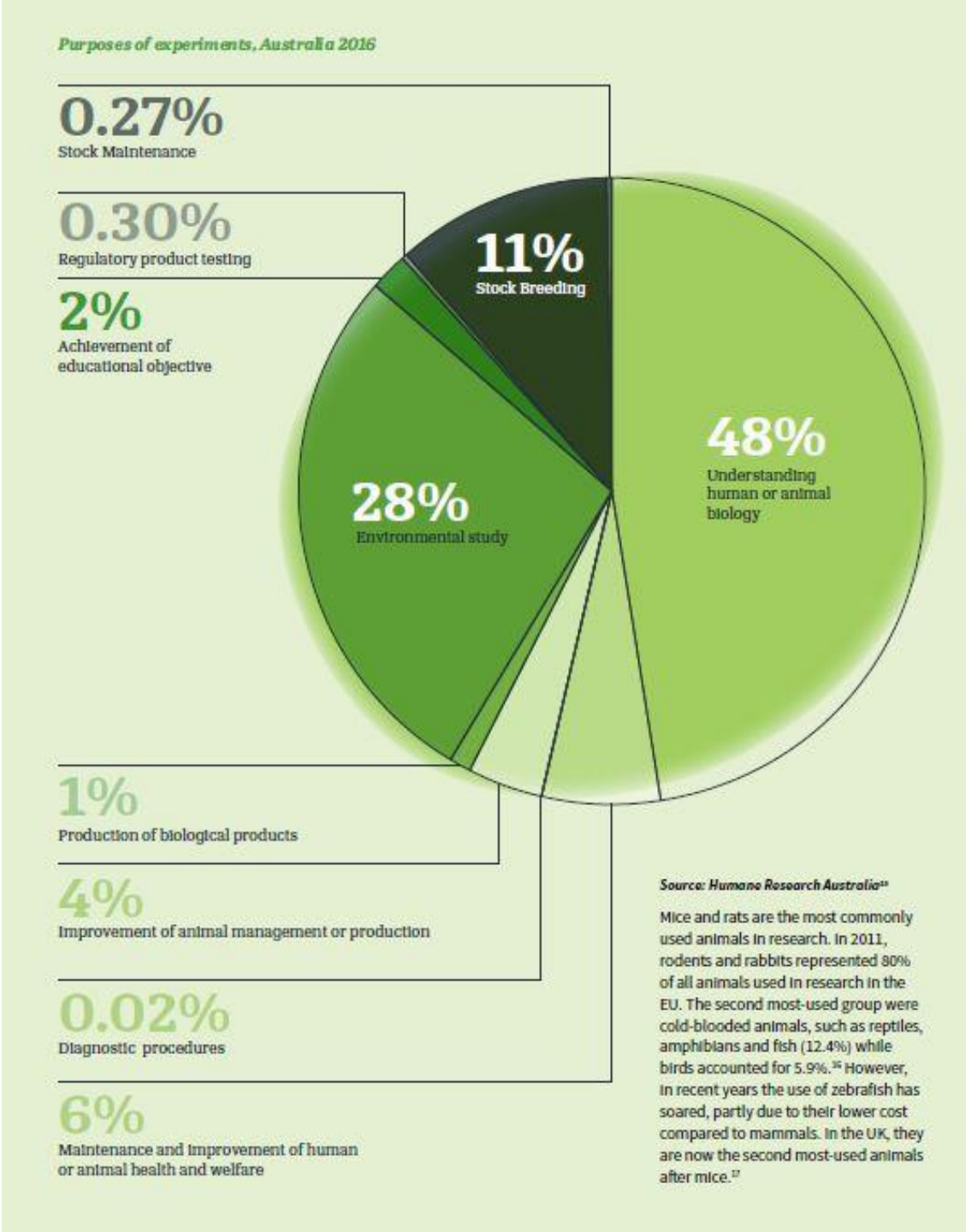
## An overview of methods and technologies that can replace animals in biomedical research and testing

Monika Merkes PhD (public health)
President, Humane Research Australia

# Extent and types of animal research

- Worldwide, more than 115 million animals are used per year to supply the biomedical industry.
- China, the US, Japan and Australia are the heaviest users.
- Animals are used in
  - Fundamental (basic) research
  - Applied (or human disease) research
  - Testing (regulatory testing)

Purpose of experiments, Australia (2016)



# How many animals are used?

- Approx. 9 million in Australia (2016)
- Just under 11.5 million in the 28 EU countries (2011)
- An estimated 115.3 million worldwide (2005)

Image source: Wikimedia Commons
https://commons.wikimedia.org/wiki/File:Lab_mouse_mg_3154.jpg

# The 3Rs

National Health and Medical Research Council (2013). **Australian code for the care and use of animals for scientific purposes**.

- Replacement
- Reduction
- Refinement

….. The 3Rs are meant to protect animals in research and testing.

# Why do we need alternatives to animal research?

- Do we have the moral right to use animals for our purposes?
- Animal research produces misleading results.
- Opportunity cost (we don’t know to what extent new drugs and treatments for humans have been overlooked, because they were harmful in animals or didn’t work in animals).
- Animal tests are costly and lengthy.

# There are now better ways …

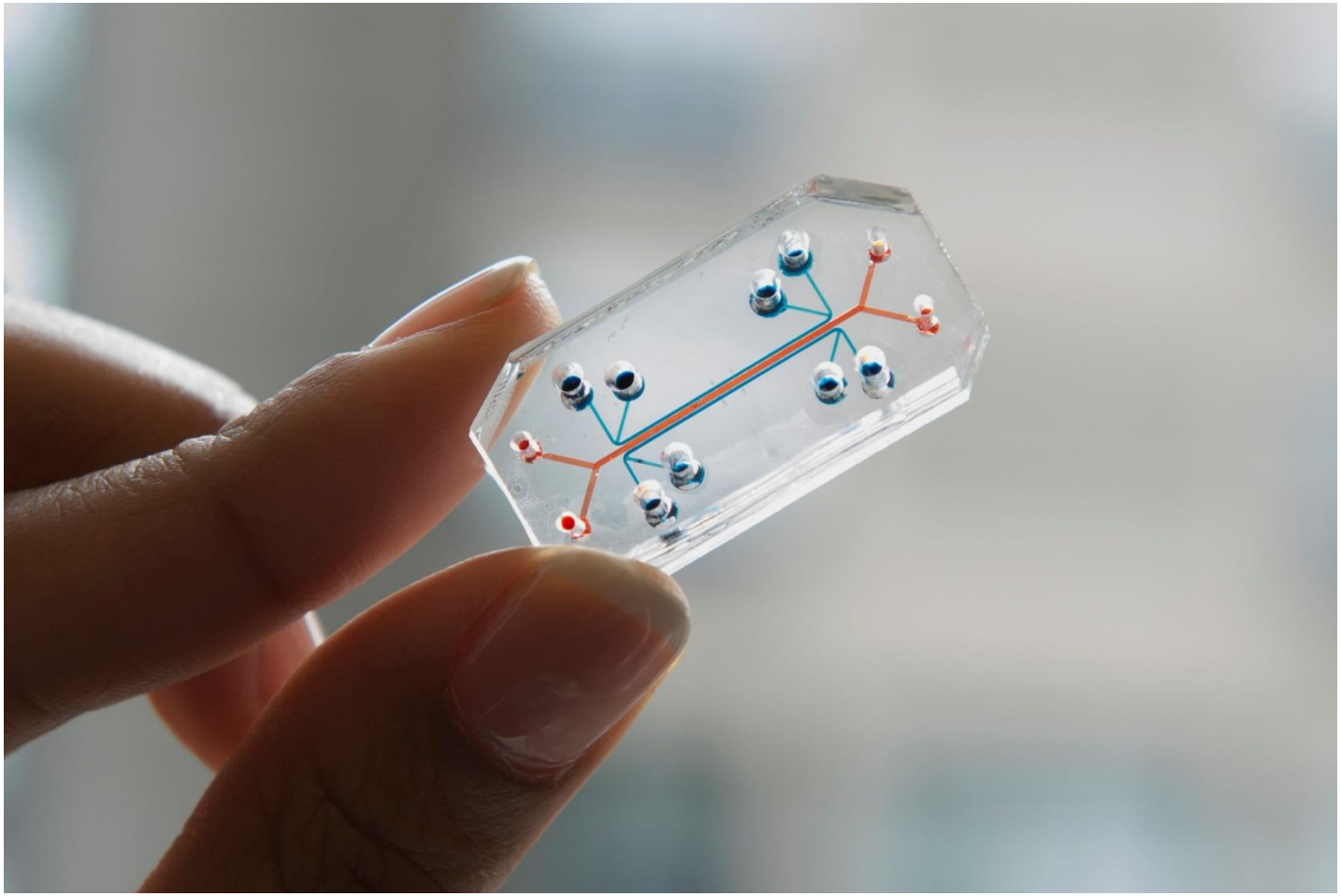
- In vitro methods (performed with microorganisms, tissues, cells, Petri dishes etc.)
- In silico (computer-based) methods
- Studies with human volunteers
- Simulators

# In vitro (test tube) methods

3D tissues and microfluidic devices:

- **Organoids** – miniature and simplified versions of an organ, grown in-vitro in 3 dimensions
- **Organs-on-chips** – mini organs can also be grown on microchips, made of a clear and flexible polymer and contain hollow microfluidic channels lined with living human cells

# Lung-on-a-chip



Source: National Center for Advancing Translational Sciences, US

# In-vitro methods

**Biobanking**

- To study cells and tissues, researchers need a readily available supply of these human biological samples. They are stored in biobanks.
- Biobanks store tissues that are left over from medical procedures, have been donated, or are collected specifically for research.

# In-vitro methods

**Omics technologies**

- Refers to areas of study in biology whose names end in “omics”, such as genomics (the study of the genome of an organism).
- Diverse technologies that focus on studies of life processes, such as studies of genes, proteins and metabolites of an organism.

# In-vitro methods

**Stem cell technologies**

- Stem cells are unspecialised or undifferentiated cells with the ability to self-renew, and to differentiate to produce specialised cell types in the body

Types:

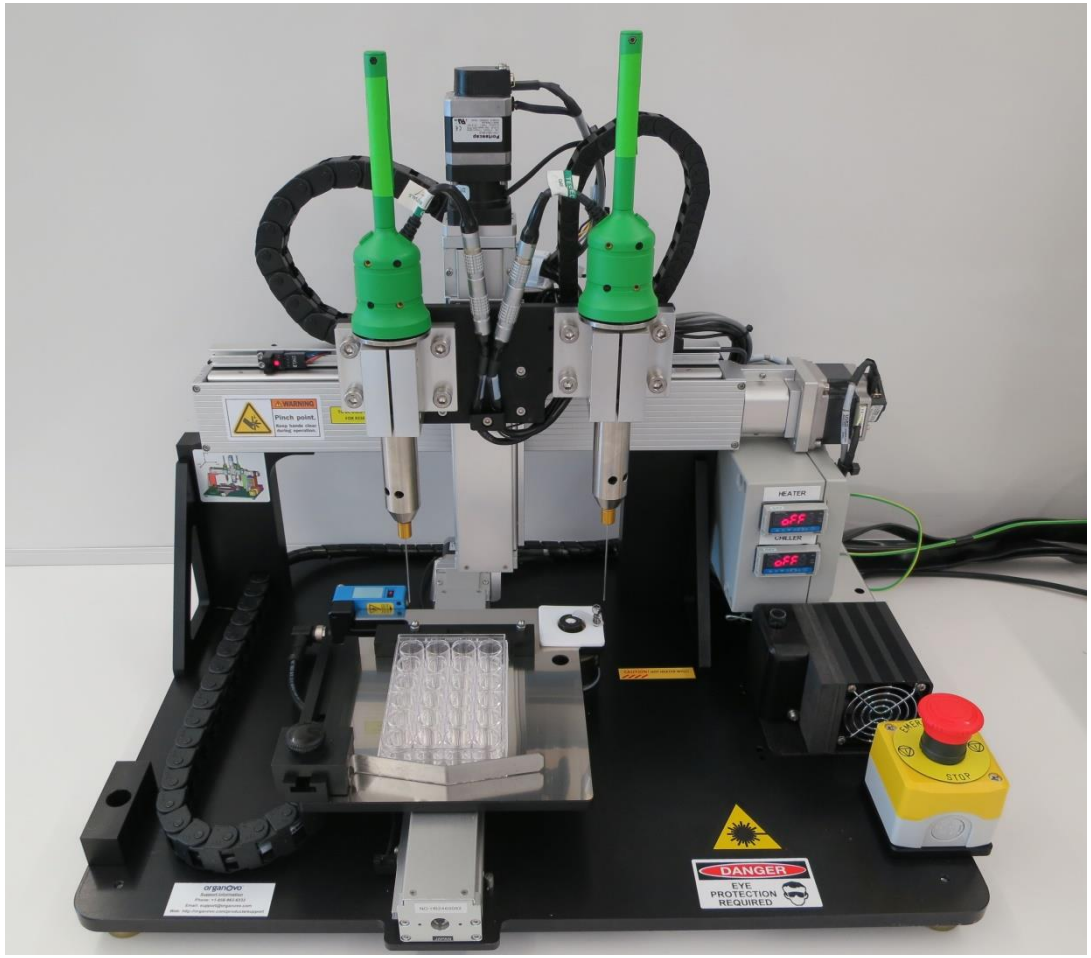
- Tissue specific stem cells (adult stem cells)
- Embryonic stem cells (5-7 days)
- Induced pluripotent stem cells (iPS cells) – mature cells can be induced to mimic the characteristics of an embryonic stem cell

# In-vitro methods

**3D and 4D bioprinting**

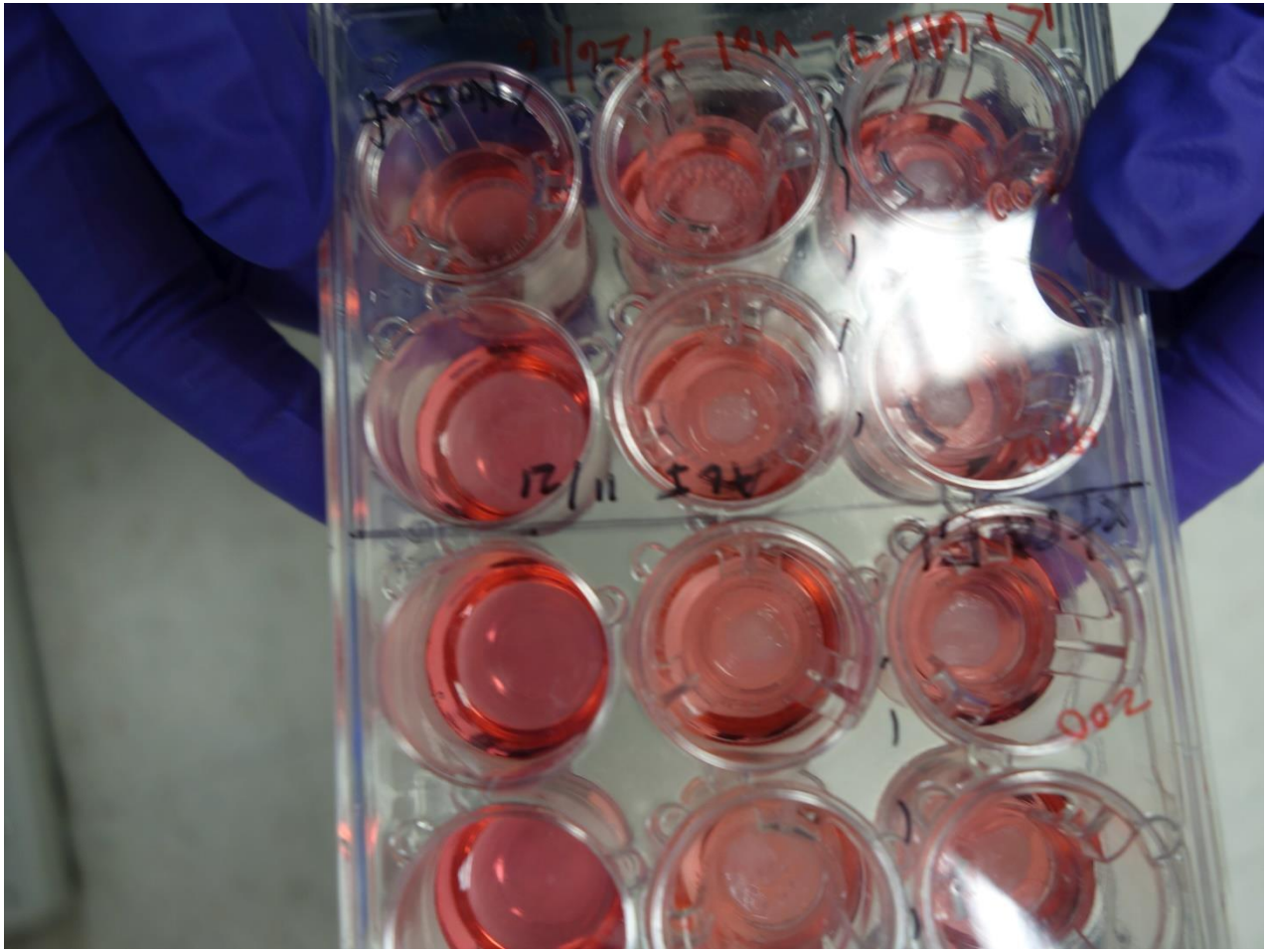
- Bioprinting involves the precise layering of cells, biologic scaffolds, and growth factors with the goal of creating bioidentical tissue for a variety of uses.
- 4D bioprinting – 3D patterned biological structures that can transform their shape or behaviours. For example, 4D printed materials can change their shape over time.

# Organovo bioprinter




Source: National Center for Advancing Translational Sciences, US

# 3D printed eye tissue

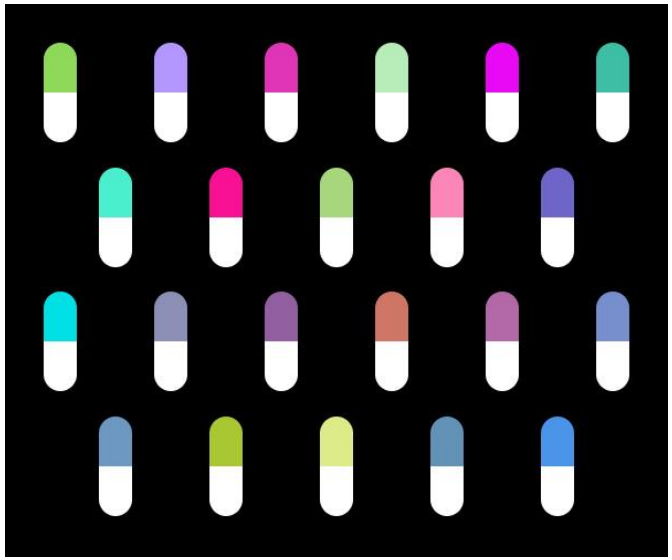


Source: National Center for Advancing Translational Sciences, US

# In-vitro methods

**Robotic tests** (high-throughput tests)

- Automated methods to test biological activities of thousands of chemicals that used to be tested in animals.



Source:
https://en.wikipedia.org/wiki/File:Abstract_pills.jpg

# High-throughput screening robots

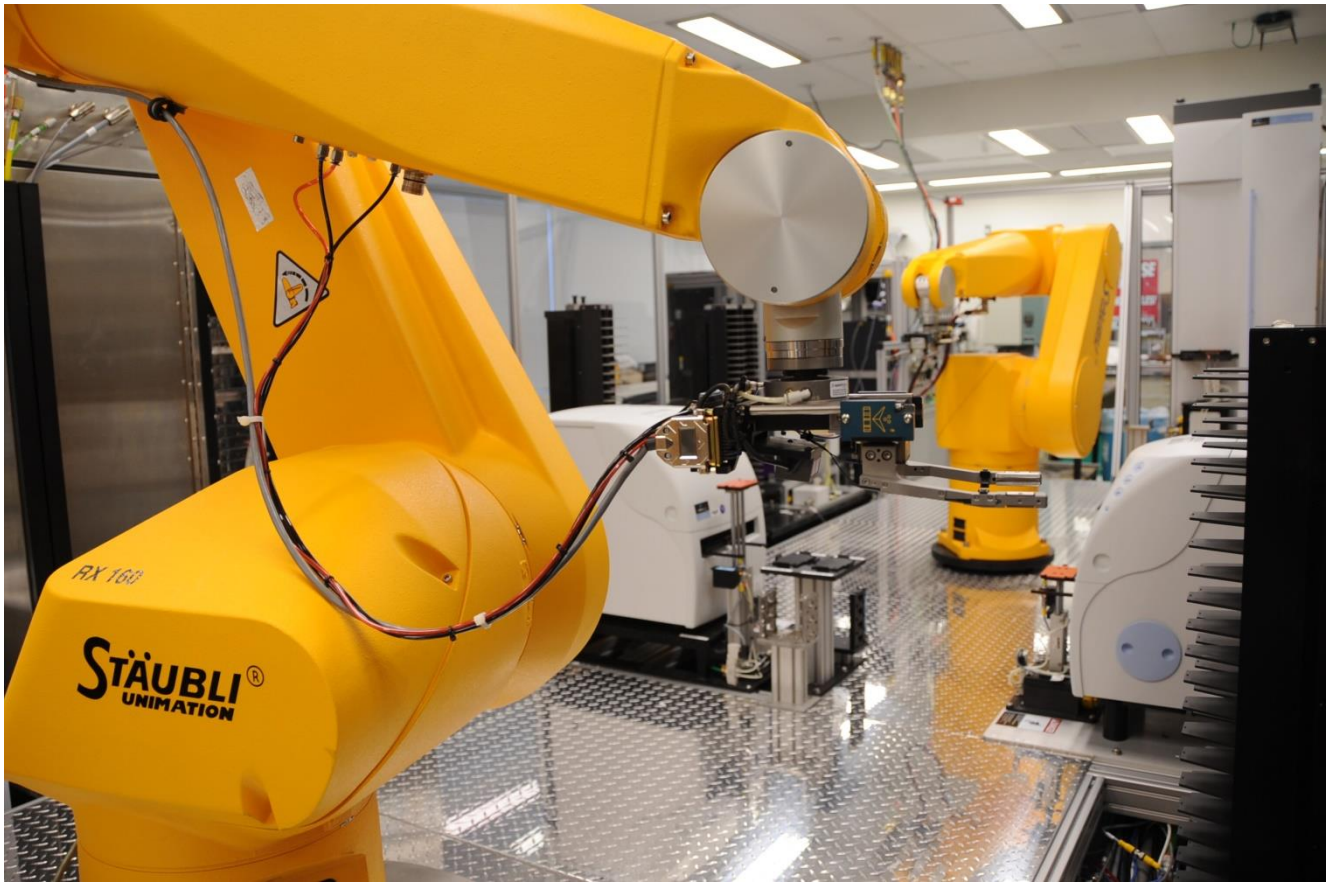
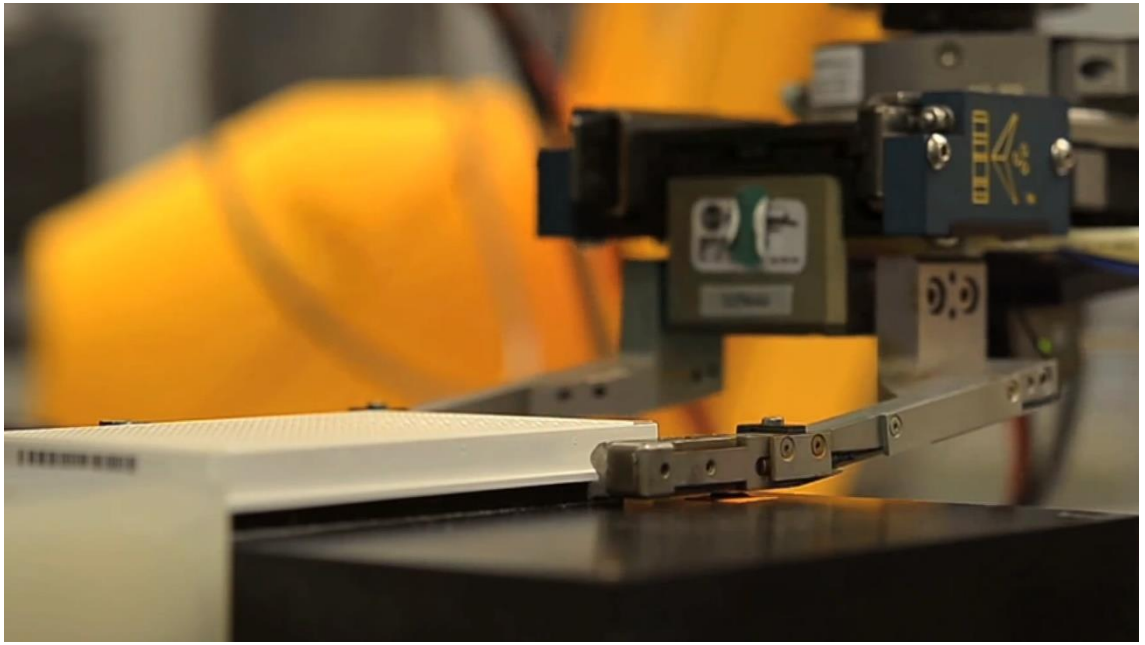



Image source: Maggie Bartlett, National Human Genome Research Institute http://www.genome.gov/dmd/img.cfm?node=Photos/Technology/Research%20laboratory&id=79299, Public Domain, https://commons.wikimedia.org/w/index.php?curid=37410189



A robot arm handles an assay plate in high-throughput screening process. This image is from a video titled "Using 3D Printing to Advance Science" created by NIAID.

Image source: National Institute of Allergy and Infectious Diseases - http://www.niaid.nih.gov/news/newsreleases/2014/Pages/3DPrintExchange.aspx, Public Domain, https://commons.wikimedia.org/w/index.php?curid=37410586

# In-silico (computer-based) methods

- Prediction methods and tools (mainly for the evaluation of toxicity)
- In-silico methods combined with other approaches (mainly for the evaluation of toxicity)
- Computer modelling

Most progress with in-silico models has been made in the area of toxicology (the study of the adverse effects of chemical substances on living organisms).

# In-silico methods

- The **read across method** uses data from a chemical substance for which safety information is available, to make predictions for a similar substance about which not much is known.
- Researchers in the US compared this new method with animal tests: read across was accurate 80-95% of the time, compared to 50-70% for animal tests.

Source: Luechtefeld, T., Marsh, D., Rowlands, C., & Hartung, T. (2018). Machine learning of toxicological big data enables read-across structure activity relationships (RASAR) outperforming animal test reproducibility. *Toxicological Sciences, 165*(1), 198-212.

# In-silico methods

**Computer modelling** of health and disease

- A computer-based model or simulation is a computer program that is designed to simulate a physical or biological system or situation (e.g., simulate virtual organs or the human body).
- Computer models can link many processes together, something which is not possible to achieve with animal models.

# Studies with human volunteers

- Post-mortem studies
- Population-based studies (epidemiology)
- Microdosing

Image source:
https://upload.wikimedia.org/wikipedia/commons/thumb/2/26/Assorted_Pills_2.JPG/1024px-Assorted_Pills_2.JPG

# Simulators

Simulators are either virtual reality (VR)-based or physical model (PM)-based. Apart from replacing live animals in education and training, VR simulators have great potential for training.


Image source: https://commons.wikimedia.org/wiki/File:PHOTOS_INSIDE_THE_CLASSROOM_UPDATED014.jpg

# Simulators

Source: Lavit Yaacov, Sheba Medical Center, Israel

# Simulators

Froggipedia


Source: App Store (iTunes)

# Efforts by governments, the scientific community & industry to replace animal experimentation

- Pooling knowledge and resources
- Working together to build large databases
- Plans for further development of technologies (such as organs-on-chip)
- Collaborating on the validation of new methods and technologies

# Concrete plans to end certain types of animal testing

US Environmental Protection Agency (EPA):

- “EPA will reduce its requests for, and funding of, mammal studies by 30% by 2025 and eliminate all mammal study requests and funding by 2035.”
- … “awarding $4.25 million to advance the research and development of alternative test methods for evaluating the safety of chemicals that will minimize, and hopefully eliminate, the need for animal testing.”

Memorandum dated 10 September 2019,
by Andrew Wheeler, Administrator, EPA

# Concrete plans to end certain types of animal testing

The Netherlands (2016):

- By 2025, animal procedures will be phased out in regulatory safety research (such as testing of chemical substances, food ingredients, pesticides, medicines and vaccines).
- It will take longer to transition to animal-free research in the fields of fundamental scientific research and applied and translational research.

# Already banned

- The EU Cosmetics Regulation prohibits animal testing of finished products since 2004 and of cosmetic ingredients since 2009
- Marketing ban of cosmetics finished products tested on animals since 2004
- Cosmetics containing ingredients tested on animals since 2013

# Australia

**Industrial Chemicals Act 2019**:

- From 1 July 2020, Australia will have a de facto ban on new animal testing for chemicals solely used in cosmetics.
- Industrial chemicals for sole use in cosmetics can be imported or manufactured only if they provide safety data that do not rely on new animal testing.

# Why does it matter?

- It has always been unethical to inflict pain and suffering on non-human animals for the purported benefit of humans.
- The “necessary evil” argument no longer holds, because of growing awareness of the shortcomings of animal experimentation and the availability of better, non-animal methods.

# Better ways to do research

PDF available from the HRA website http://www.humaneresearch.org.au/

For a hard copy, ring HRA on 1800 486 263