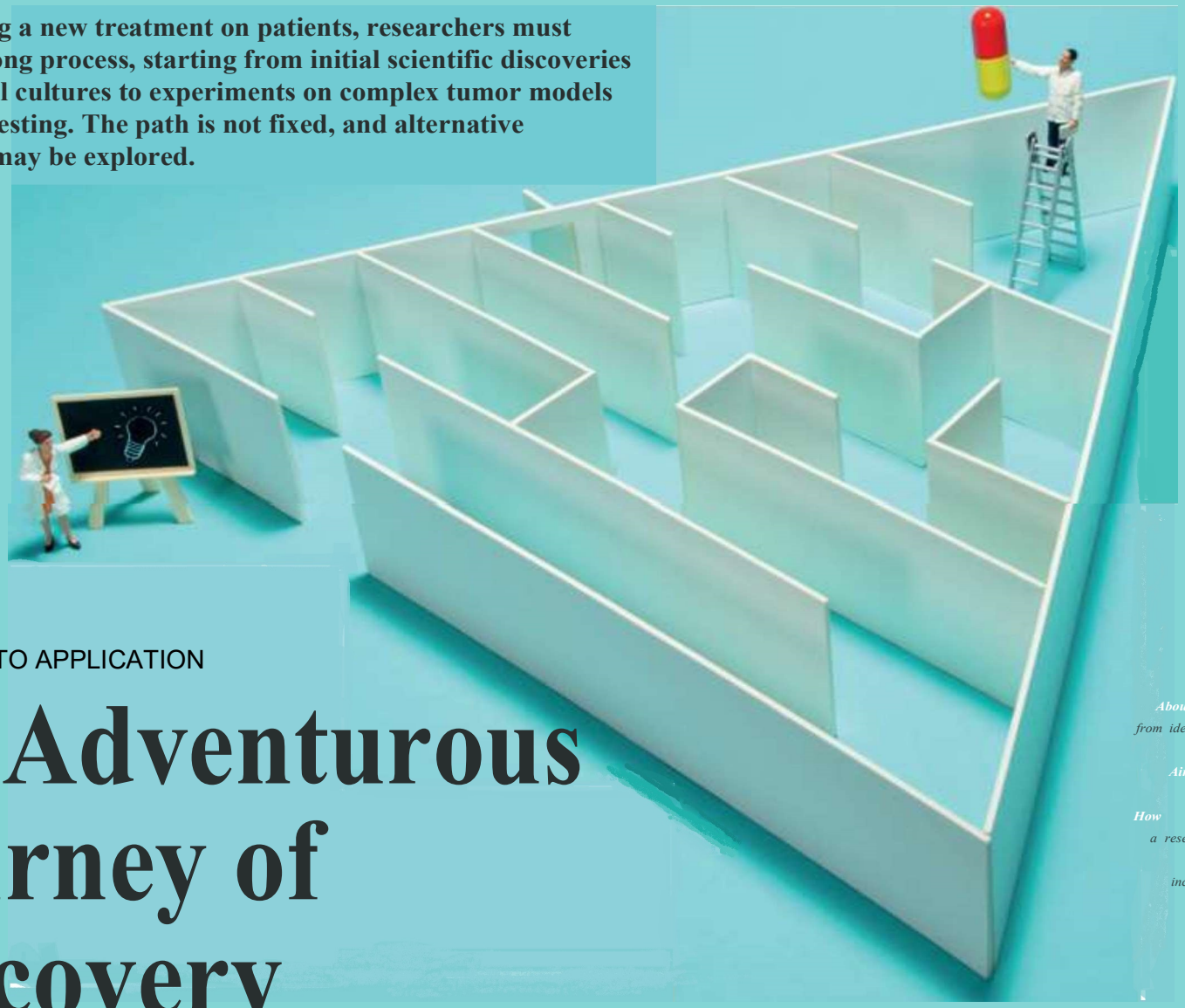
Before testing a new treatment on patients, researchers must complete a long process, starting from initial scientific discoveries on simple cell cultures to experiments on complex tumor models and animal testing. The path is not fixed, and alternative approaches may be explored.



FROM IDEA TO APPLICATION

An Adventurous Journey of Discovery

About *The long road from idea to application in patients.*

Aim *Developing new cancer treatments*

How *By formulating a research question and testing it with increasingly complex experiments.*

All scientific research at Antoni van Leeuwenhoek shares a common goal: improving cancer treatment, including developing new drugs. The process is lengthy, beginning with an idea and progressing through laboratory experiments, from simple to highly complex. Many promising ideas are discarded due to disappointing results, ineffectiveness in complex environments, or excessive harm to healthy tissue. Only the most successful treatments that pass all prior tests are eventually tested on humans, potentially leading to new therapies.

THE RESEARCH QUESTION

Eureka? The image of the scientist coming up with a brilliant idea in their bathtub is long outdated. In reality, the research world resembles an anthill where people continuously inspire each other with ideas, insights, and surprising research questions. It's no coincidence that scientific articles sometimes list up to thirty authors. Science is a team sport. A new research question can arise from a discussion at a conference or during a meeting, or from a new technology. Patient treatment can also lead to fundamental questions. Once research has started from a question that has arisen, it will often still go in a completely different direction, either because another direction proves more promising or because more knowledge about something else is needed first. Scientists describe their profession as an exciting journey of discovery where you can end up somewhere completely different than you expected.

PYRAMIDE

Various methods are available to answer research questions, and the number continues to grow. You can view laboratory research as a pyramid, with the simplest research at the bottom, more complex experimental models above, and animal testing at the very top. There is no fixed route: everything depends on the research question.

Research can start, for example, with a cell line: standardized cancer cells in a petri dish, available for purchase from a manufacturer. Cell lines can be used to check whether a particular drug inhibits the growth of cancer cells. They are simple to use and relatively inexpensive in terms of time and money. A disadvantage is that the behavior of a cell line bears little resemblance to the behavior of a cancer cell in an actual patient. If a drug works on a cell line, it offers no guarantees. Another possible starting point is a computer model, which simulates biological or chemical reactions without laboratory experiments.

INCREASINGLY COMPLEX MODELS

After the initial experiments, more advanced models are used to more closely simulate a patient's tumor. These include cell lines from a fresh tumor sample or a 3D cell culture of the patient's tumor. If the drug remains promising, further tests involve tumor organoids, tumor avatars, or an organ-on-a-chip (see interviews, red.), a small glass device with channels and spaces where human cells grow.
If a new treatment successfully passes many of these tests, it proceeds to animal testing, a step legally required before human trials: a treatment not tested

on mammals cannot be researched in humans. If animal testing shows favorable results, further analyses by certified companies follow. This also ensures the objectivity of the research. Only after approval by competent authorities, such as the EMEA, can the first patients start with an experimental treatment. This is when clinical research begins.

Prof Jos Jonkers about tumor-organoids

'New medicines can be tested more effectively'

A tumor organoid consists of three-dimensional cultured cells that grow from a patient's tumor cells. 'Since the 1950s, we have been able to grow cells in the lab, but until recently, only on a flat surface,' says researcher Prof. Jos Jonkers. One advantage of three-dimensional cultures is that you get more different types of cells together, just like in a real tumor in a patient. A tumor organoid is suitable, among other things, for testing new therapies. 'With the previously used cell cultures, it often happened that a medicine worked very well in the lab, but not at all in patients. We hope that organoids can better predict effectiveness. We are not sure yet, as these organoids have only existed for ten years.'

Researchers are keen to establish biobanks of tumor organoids.

If it is possible to culture tumor organoids from the cells of a large number of patients, for example, those with colon cancer, you would have a kind of surrogate group of comparable patients to test new therapies on. This offers advantages. For instance, a medicine might only be effective with tumor organoids from patients With certain characteristics. Then you can focus subsequent research specifically on that patient group.' Tumor organoids also have their limitations. 'For example, they do not contain immune cells. The environment of healthy cells is missing, and also blood vessels are absent."

More refined models are being developed to incorporate these elements. For example, you can grow tumor organoids in a test animal instead of in a gel full of nutrients and growth factors. This way, you have the environment of healthy cells and blood vessels you can then, for example, see if a medicine is well absorbed and reaches the tumor. However, the human immune system is still missing, although there are possible solutions for that as well.' Jonkers is enthusiastic about the ever-expanding range of experimental models in the lab. 'You can test new medicines more effectively before starting research on patients. And this is urgently needed, because currently, only five percent of all new treatments that reach that stage are successful. By using all research models, from cell lines to tumor organoids and test animals, we hope to double that percentage. But for that, you will still need the entire range of experimental models; tumor organoids alone won't suffice.'

Dr. Els Hermans en dr. Marieke van de Ven over de rol van dierproeven

'No research model comes close to a living organism, yet'

Mandatory animal testing before human trials is subject to strict rules and regulations. A medical ethics review committee must approve them in advance. Animal experiments can only be conducted if the research question cannot be answered in any other way.
Is it not possible to skip animal testing now that refined models, such as,tumor-organoiden and tumor-avatars have been developed? 'No, unfortunately not,' Dr. Els Hermans, head of the Animal Testing department, explains that however these models are valuable, they cannot fully replace animal models. 'I understand that people think that way as the media often features many hopeful quotes. However, while complementary models are important, they are not yet sufficient. A tumor organoid or tumor avatar cannot replace an animal model. No model yet comes close to a living organism with blood vessels, immune cells, connective tissue, nerve cells, and so on. For certain research, such as how a tumor metastasizes or the long-term effects of a treatment, animal testing will likely always be necessary. A tumor-organoid or a tumor avatar may have blood vessels however the blood vessels in a tumor avatar for example can survive only a couple of days, and serves a different function than a living organism. The process of metastasis, and long-term effects of treatments can not be studied in such a model. Therefore, animal research will still be necessary'.
Marieke van de Ven, head of the Mouse Cancer Clinic, cites immunotherapy as an example of a treatment that would never have come about without animal testing. 'To effectively combat a tumor, you need an immune system capable of responding swiftly. This requires a complete organism, as a mere clump of cells is insufficient. Immunotherapies can indeed be tested in tumor avatars, but these models cannot be used to observe long-term responses and the development of resistance. Moreover, animal testing is not only necessary for developing new medicines and studying side effects, but also for better understanding the biology of cancer. Ultimately, this has the same goal: to improve cancer treatment by finding new targets for therapeutic interventions.'

Daniela Thommen about tumor-avatars

‘Everything that is present in the original tumor, is also in the tumor avatar’

Dr. Daniela Thommen and her research group are developing models specifically designed for immunotherapy research. ‘Immunotherapy does not target cancer cells directly, like chemotherapy or radiation, but activates the immune system to fight the cancer,’ she explains. ‘It is a major breakthrough: some immunotherapies work wonderfully. But often only in at most ten to fifteen percent of patients. Researchers therefore want to develop new immunotherapies. The problem is that there are too few patients to test all these new therapies on. And we also cannot yet identify the right patients who could benefit from them.’ Thommen is therefore trying to find out how immunotherapy precisely affects the biology of the tumor. This knowledge should help to select the right patient groups and to develop more rationally based immunotherapies. ‘Mouse models are very good for researching immunotherapy,’ says Thommen. ‘Blood vessels, lymph nodes: everything is included. Nowadays, tumors can even grow in a “natural” way thanks to genetic mutations in the mice. But to study all the possible genetic variations that occur in patients, you would need to study hundreds of different mice. Tumor organoids grow in a more natural way, but it is not possible to maintain the architecture and complexity of tumors in patients.’ Thommen developed a new research model that nicely complements the other models: tumor avatars. ‘With the patient’s permission, we receive small pieces of tumor tissue left over after surgery. We cut these into fragments of about 1 cubic millimeter. Then we culture them, after which we can treat them with immunotherapy. The great advantage is that everything that was in the original tumor is also in the tumor avatar - including immune cells. Another advantage is that you can compare different immunotherapies on the tumor avatars, something that is not possible with patients. We can often get many pieces from a small piece of tissue. We can freeze these and later thaw them to try a new combination. Thommen and colleagues have already shown that the response of the tumor avatars to a commonly used immunotherapy corresponds well with the response of the patient from whom the tumor tissue was taken. ‘This gives confidence in the predictive value of this new research model. However, tumor avatars also have their limitations. For example, they do not contain blood vessels or lymph nodes that play a role in the immune response of patients. Unlike organoids, you cannot expand tumor avatars; they survive only a few days in culture. You also cannot study the effects on other organs and tissues. For that, test animals are still needed. But thanks to tumor avatars and other new research methods, we can use test animals more selectively.’