Novel use of hormone receptors

How to Bomb Cancer?

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Most peptide hormones bind to specific molecules on a cell's surface. Biotechnology offers some clues about how we can use this "key-lock" interaction to defeat cancer

Over 100 years ago the German scientist Paul Ehrlich envisaged the concept of a "magic bullet". It would be able to find and kill cancer cells by identifying the specific particles (antigens) present on their surface and leaving healthy tissues intact. This idea remained essentially unexplored for many decades. We hope that our contribution will help to finally make it come true.

There has recently been growing interest in combating tumors by mimicking the activity of the immune system. The technique of combining antibodies with toxins promises to make it possible to deliver such toxins selectively to mutated cells. Antibodies would first recognize specific antigens and settle down on cancer cells, and then the bound toxins would eradicate the cells. Antibodies coupled to radioisotopes such as iodine 131, iridium 90 and many others are also being tested in the treatment of cancers.

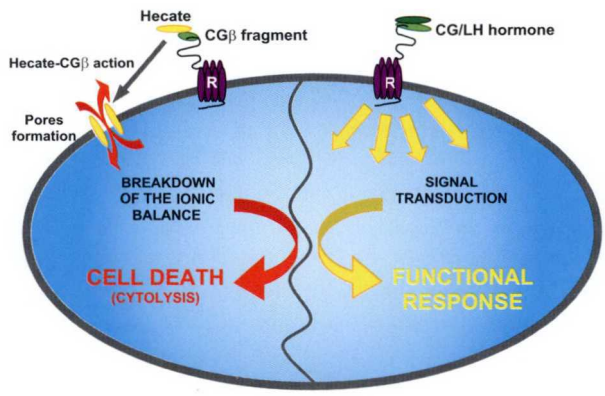
Unfortunately compounds based on antibody selectivity turned out to work less efficiently than expected, mainly due to the instability of toxin-antibody binding or because of negative patient response to toxins or monoclonal antibodies, which complicated long-term therapy. In the 1980's, conjugates of toxic compounds with hormones were applied. Hormones were used as carriers of toxins for eliminating prostate and mammary gland cancer cells.

One promising group of toxins is called "defensins" - lytic peptides that are widely prevalent throughout the animal and plant kingdoms. Defensins are an evolutionarily ancient weapon, which scientist have only been exploring for the last 20 years. Today we have identified about 500 defensins that are able to fend off a wide range of bacteria, protozoa, fungi and viruses containing cellular membrane. We suspect that they kill cells by binding to the negatively charged cellular membrane, creating holes that cause the contents to leak out, leading to cell death. However, there is one problem with using them as prospective pharmaceuticals: when released into the blood, defensins become rapidly deactivated through enzymatic degradation or through non-specific binding to plasma elements.

Protein of fate and revenge

Conjugating such antimicrobial lytic peptides with units that target them to tumors prolongs their period of action. Furthermore, such a combination reactivates the apoptosis (programmed suicide) of cancer cells, which ceases to function properly during the cancer transformation. Taking this into account, a conjugate called "Hecate-CGβ" was synthesized, named after Hecate, a goddess of human fate, magic and revenge in Ancient Greek mythology. Scientists hoped that this peptide would demonstrate the talents of its namesake. In our research, Hecate-CGβ is a fusion of the lytic peptide Hecate (a synthetic analogue of a major component of bee's venom) and a short fragment of the hCG (human chorionic gonadotropin) hormone. This part of the hCG hormone acts as a key that carries the toxin to the lock-receptor of luteinizing hormone (LH). Besides being present in the ovaries and testes, LH receptors are found in many other tissues, i. e. the uterus, mammary glands, the prostate or tumors derived from these organs. This makes it possible to cause the targeted ablation (eradication) of cancer cells: after the CGβ recognizes its lock, Hecate comes into action, working its "black magic" and killing the cell.

At our Institute's Department of Hormone Action Mechanisms, we have managed to confirm this mechanism for the first time, by demonstrating the characteristics of Hecate-CGβ binding to membrane LH receptors. In cooperation with Prof. W. Hansel (Louisiana State University, USA) and



By binding to the membrane receptor, the lytic peptide (Hecate)-hormone (CG) conjugate (left) interrupts the normal cell function (right)

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Laboratory rats were the first to benefit from the unexpected influence of toxin-hormone conjugate.

the group of Prof. I. Huhatniemi (University of Turku, Finland), we have shown that the selective ablation of cancer cells can be achieved using the Hecate-CGβ conjugate. In very low concentrations it caused the death of cancer cells in the ovary, testis, prostate or mammary gland, and its lytic activity was directly correlated with the number of LH receptors. Dr. Gabriel Bodek investigated the effect of *in vivo* Hecate-CGβ on transgenic mice with developed ovarian or testicular tumors. After 3 injections (once a week), a 60% and 36% decrease in testicular and ovarian tumor size, respectively, was found, although the spectacular remission of ovarian tumors was even observed in some females.

Unexpected mechanism

Encouraged by promising results of *in vitro* studies performed on various cells as well as by using dies on transgenic mice, we applied Hecate-CGβ to the treatment of prenatally induced mammary gland tumors in rats. The conjugate appeared to be very effective, clearly limiting growth of expanded tumors as compared to nontreated (control) rats treated with Hecate peptide alone. Our astonishment was all the greater when we confirmed the presence of LH receptors in the mammary glands of only 17% of the experimental animals. Surprisingly, we found that Hecate-CGβ treatment drastically decreased the concentration of growth hormone and prolactin in circulating blood. Additionally, a significant decline in growth factor content was also detected in cancer tissues. These results suggest that Hecate-CGβ affects rat mammary gland tumors by either a direct or indirect mechanism. The direct action could involve the preferential disruption of cancer cells, which, unlike healthy eukaryotic membranes, maintain large membrane potentials similar to prokaryotic cells. This known phenomenon could be involved in the regression of experimental mammary tumor growth. On the other hand, an indirect mechanism is also possible: Hecate-CGβ causes decreased secretion of tropic metabolic hormones, creating an unfavorable environment for tumor growth.

Although the precise mechanism by which the Hecate--CGβ conjugate functions is not clear, we were able to ascertain that this compound has the potential for use in the *in vivo* treatment of animal ovarian, testicular and mammary gland tumors. Perhaps further investigation will lead us to a treatment for certain cancers in mammals - including in humans. ■

Further reading:

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