amoeboid parasite *Entamoeba histolytica*[13] and the pathogen *Trachipleistophora hominis*[14] (which belongs to a group of fungi called microsporidia). Before those reports, *Entamoeba* and microsporidia were thought to lack mitochondria altogether. Now mitosomes appear in *Giardia*, too, and we have the first direct glimpse of their function.

The biochemical role of the mitosome can hardly be core ATP synthesis. That occurs in the cytosol in both *Entamoeba*[5] and *Giardia*[5]. And it is uncertain that microsporidia even make their own ATP, because they can steal it from the cells that they infect[15]. Tovar *et al.*[4] show that, instead, the *Giardia* mitosomes harbour critical enzymes of Fe–S cluster assembly. Furthermore, cell fractions enriched for the organelle assemble Fe–S clusters *in vitro*. This pins a function to the mitosome in *Giardia* and is a major advance in understanding.

Possibly, *Giardia* assembles Fe–S clusters in mitosomes for the same reason that yeast and humans assemble them in mitochondria: the process is very oxygen-sensitive. The mitochondrial matrix (the space inside the organelle) is the most oxygen-poor compartment in oxygen-respiring cells because oxygen is consumed in the surrounding membrane. Lloyd *et al.*[16] have shown that *Giardia* possesses organelles that accumulate mitochondrion-specific dyes and that can transfer electrons from donors to acceptors, which could easily include oxygen. But it remains to be seen whether the organelles observed by Lloyd *et al.*[16] and Tovar *et al.*[4] are identical.

Although mitochondria are usually considered to be oxygen-dependent, *Giardia*'s tiny mitochondria have an anaerobic function (Fe–S cluster assembly) in the synthesis of oxygen-sensitive proteins such as hydrogenase and PFO, which are normally found in hydrogenosomes[1,4–9,12]. Eukaryotes diversified while the oceans were largely anoxic[17,18], so these anaerobic functions are most easily seen as biochemical relicts of the mitochondrion's anaerobic past. Such anaerobic relicts are abundantly preserved in diverse eukaryotic lineages today[9,12].

We know that mitochondria arose as intracellular symbionts in the evolutionary past[11]. But in what sort of host? That question still has biologists dumbfounded[1]. In the most popular theories, *Giardia* is seen as a direct descendant of a hypothetical eukaryotic host lineage that existed before mitochondria did[2,3]. But Tovar and colleagues' findings[4] show that *Giardia* cannot have descended directly from such a host, because *Giardia* has mitosomes. So our understanding of the original mitochondrial host is not improved by these new findings, but our understanding of mitochondria certainly is. In its role as a living fossil from the time of prokaryote-to-eukaryote transition, *Giardia* is now retired. But it assumes a new place in the textbooks as an exemplary eukaryote with tiny mitochondria that have a tenacious grip on an essential — and anaerobic — biochemical pathway. ■

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Chemistry

Dendrimers set to self-destruct

E. W. Meijer and M. H. P. van Genderen

The versatility of the branched macromolecules known as dendrimers is being exploited in various ways — explosively so, in the context of their application as potential drug-delivery systems.

If a good idea for scientific innovation emerges, you can be sure that several teams of researchers will be quickly on the case. An example comes in the form of three reports[1–3] that explore the prospects of using dendrimers for drug release inside diseased cells.

Dendrimers are artificial macromolecules, constructed in step-by-step fashion using repetitive chemistry. The macromolecule constituents radiate in branching form from a central core, creating a sphere of chemical groups that can be tailored according to requirements. The results of the process are not only aesthetically appealing but offer chemists wonderful opportunities for exploring new ideas[4]. Dendrimers are large, but can be synthesized and characterized with a precision similar to that possible with smaller organic molecules. They do not suffer from the problem of 'polydispersity' that dogs linear macromolecules: that is, constituents of a given set of dendrimers have exactly the same molecular weight, rather than being a mixture of chains with a distribution of molecular weights. And the large number of identical chemical units in the branching units, as well as those at the periphery, confers great versatility. The end groups can be designed for various purposes, including sensing, catalysis or biochemical activity.

In this last instance, one of the potential virtues of dendrimers comes under the heading of 'multivalency': the enhanced effect that stems from lots of identical molecules being present at the same time and place. The combination of multivalency with precision architectures has made dendrimers of increasing interest for biomedical applications, not least for drug delivery[5]. Dendrimers can enter cells remarkably easily, a property that means they have been investigated as potential gene-transfection agents. The chemical groups that bristle from the ends of the branches allow for tuning of biological properties, and can anchor one or more target groups onto the dendrimer. The compound that constitutes the drug itself can be physically encapsulated in the dendrimer or bound to it. There have been attempts to achieve total and simultaneous release of active agents through changing pH conditions. But generally the traditional route has been that of getting one chemical trigger to release one drug molecule.

Independently of one another, teams led by de Groot[1], Shabat[2] and McGrath[3] have explored a much more advanced concept — simultaneous release of all of a dendrimer's functional groups by a single chemical trigger. All three exploit the fact that the dendrimer skeleton can be constructed in such a way that it can be made to disintegrate into known molecular fragments once the disintegration process has been initiated. Variously termed "cascade-release dendrimers"[1], "dendrimer disassembly"[3] and — colourfully — "self-immolative dendrimers"[2], these systems in effect perform a chemical amplification reaction. Triggered by a specific chemical signal, the dendrimer scaffold falls apart in several steps in a chain reaction, releasing all of the constituent molecules.

Two of the teams[2,3] demonstrate the process in systems in which relatively simple

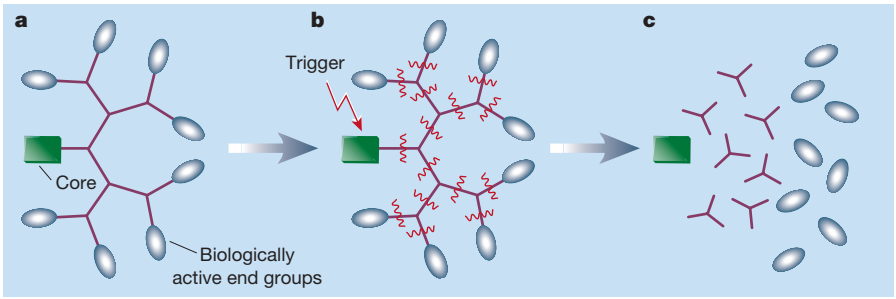

Figure 1 **Bang on — a simplified depiction of the simultaneous release of biologically active end groups from a dendrimer[1–3].** a, **The basic dendrimer, shown here as a two-dimensional part of a sphere.** b, **Triggered by a specific signal, the dendrimer scaffold falls apart in a chain reaction.** c, **The result is release of all the constituent molecules, including the end groups. In the experiments of de Groot *et al.*[1], these end groups consisted of molecules of the anticancer drug paclitaxel (Taxol).**

molecular fragments are released. De Groot and colleagues[1], however, have applied the principle in an especially elegant and appropriate way. They have not only devised methods for releasing the anticancer drug paclitaxel (Taxol), but also show that the dendrimer degradation products are not cytotoxic — except for paclitaxel itself, of course, which has the job of killing cancerous cells.

The first reaction activates the dendrimer core, initiating a cascade of 'elimination' reactions that lead to drug release (Fig. 1). Biodegradable polymers have been used before as drug carriers. But because dendrimers are so well defined, they allow fine control of the size, shape and composition of the release system. Their dendritic form, with many identical units, means that amplification can be achieved as a kind of explosion. A possible drawback, however, is the same that applies to every bomb — if the trigger is activated at the wrong time or place, the result will be devastating.

The concept described in the three papers[1–3] is intriguing, but will obviously need much more development before it can be put into practice in living cells. The next hurdle to overcome will be identifying a fuse that can be ignited to act as the trigger in biologically relevant conditions. Enzymatic reactions seem a promising avenue to explore. The pay-off of this approach, if it proves feasible, would be dendrimers that are specific for the enzymes present only in the cells to be targeted by a particular drug. ■

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Virology

Fresh assault on hepatitis C

Charles M. Rice

Hepatitis C virus causes severe liver disease. Initial trials of a newly developed agent that prevents the virus reproducing itself look promising. But what are the future prospects for this treatment?

As researchers gradually got to grips with viruses that attack the liver, two were identified early on — hepatitis A and hepatitis B. But clearly there were more, because there were also cases of hepatitis that had the hallmarks of neither virus. These cases occurred following blood transfusion, and for some time the resulting disease was known as non-A, non-B post-transfusion hepatitis. The culprit, hepatitis C virus (HCV), was identified in the late 1980s. Now, nearly 15 years later, the first small-molecule inhibitors of HCV are being tested in humans. On page 186 of this issue[1], Lamarre *et al.* report spectacular success — at least in short-term trials — with an orally administered inhibitor of a viral protease, an enzyme that is essential for HCV to reproduce itself.

Hepatitis C is a severe medical problem. After the identity of the virus was revealed, the advent of reliable diagnostic methods led to the realization that here was a global infection afflicting some 170 million people. It is transmitted by contaminated blood, fortunately now rare in transfusions. Chronic — persistent — infection is typical, and the disease course is unpredictable: some people seem to be unaffected, whereas others develop cirrhosis, end-stage liver disease and liver cancer. For many, a liver transplant is the only hope.

The virus itself has a tiny genome, consisting of a single-stranded RNA molecule only about 10,000 bases in length. This RNA serves not only as the template for viral replication, but as the viral messenger RNA for further viral production. It is translated into a long 'polyprotein', which is chopped up by cellular and viral proteases into at least 10 proteins that participate in RNA replication and new virus assembly.

One would think that this so-called simple virus would be easy to defeat. Not so. HCV has been remarkably adept at frustrating not just the human immune system, but also drug developers and virologists. It is not easy to study, in cell culture or in other animals. The virus replicates poorly in cell culture. Animal infection models are limited to the chimpanzee[2], or immunodeficient mice carrying engrafted human liver cells[3]. Only recently has it become possible to study how the RNA replicates[4,5], and how HCV enters cells[6,7], in cell culture.

The human immunodeficiency virus also has an RNA genome, which for obvious reasons has been the subject of intensive investigation. Following the success of protease inhibitors in suppressing HIV replication, the HCV-encoded serine protease emerged as a favourite target in the race for new anti-HCV drugs (the 'serine', incidentally, refers to a critical amino acid at the enzyme's active site). The enzyme itself resides towards one end, in the amino-terminal third, of a protein called NS3, and it cleaves the polyprotein at four downstream sites. No one knows precisely why, but the protease is essential for HCV replication[8].

In early work, the protease was purified and its substrate specificity determined, and those in the field anxiously awaited determination of its structure. This was first reported[9,10] in late 1996. But to the dismay of the drug designers and medicinal chemists who use structure as a guide, the substrate-binding channel of the protease was shallow and relatively featureless. It lacked the nooks, crannies and pockets that are used to select and refine highly specific and potent inhibitors. Many companies moved on, focusing on other HCV enzymes.

A few hardy groups pressed on, however, including Lamarre and his team. Key to the development of their successful antiviral agent, BILN 2061, was the observation that peptides mimicking amino-terminal cleavage products are competitive inhibitors[11,12]. Starting with a weak enzyme inhibitor consisting of six amino acids — a hexapeptide — they crafted highly potent and specific hexapeptide inhibitors. They then trimmed the