Are Golgi satellites the key to understanding nicotine addiction?

- Kicking a smoking habit is difficult, but why?
- To answer this pressing question, Professor William N Green and fellow researchers at the University of Chicago and elsewhere in the USA focus on elucidating nicotine addiction and the working of smoking cessation drugs.
- Their continued research has led them to single out organelles in our cells, called Golgi satellites.
- The researchers have discovered the significant role of Golgi satellites in the upregulation of specific nicotinic receptors.
- The progression of their research equips the scientific community to design new smoking cessation therapies.

Globally, smoking tobacco is one of the leading causes of preventable death. The World Health Organization estimates that more people die every year from tobacco use than HIV, tuberculosis, and malaria combined. Why do so many people struggle to stop smoking? The main reason is that smoking is highly addictive, but we do not know much about the mechanisms underlying this addiction. Through a series of studies, William N Green, Professor of Neurobiology at the University of Chicago, USA, and colleagues have significantly added to our comprehension of the biology of nicotine addiction and how smoking cessation drugs work. Their research paves the way for developing novel smoking cessation drugs.

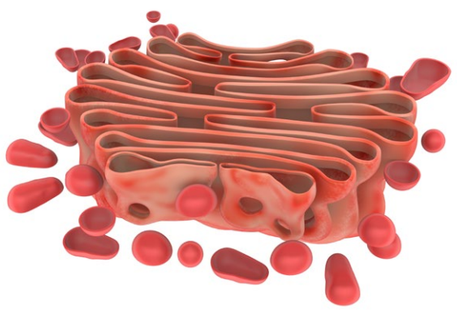
Nicotine, tobacco's addictive compound, binds to specific receptors on neurons in the brain called nicotinic acetylcholine receptors (nAChRs). These protein receptors are known as 'ligand-gated ion channels' because they open to allow ions to pass through the cell membrane in response to a ligand (a chemical messenger). In doing so, they transmit signals needed for cellular and brain function. These receptors consist of different subunits, most commonly the α4 and β2 subunits. The primary receptors implicated in nicotine addiction are α4β2Rs receptors. One effect of nicotine binding is the release of dopamine – the 'feel-good hormone' – from the reward centre of the brain. However, a person's consistent exposure to nicotine makes the receptors less sensitive for it. To compensate for this change, the receptors are upregulated by modifications that increase their function and alter other receptor properties.

This α4β2R upregulation is believed to be central to the mechanism of nicotine addiction.

The rise and fall of nicotine

People smoke intermittently, meaning nicotine levels are not constant, first rising and then falling with each cigarette. However, over the course of a day, nicotine levels in the blood will consistently rise until they level out, as displayed in Figure 1, which shows the shape of this relationship to be much like a hill with a plateau at the top. Nicotine crossing the blood–brain barrier into the brain can access all nAChRs on neurons. The consistent level of nicotine causes functional changes in these nAChRs, with dopamine release likely increasing a person's craving for nicotine.

When a person stops smoking for the day – usually before sleeping – blood nicotine levels drop rapidly, but α4β2Rs remain functionally upregulated longer than 24 hours. Subsequent decreases in dopamine are thought to amplify



The Golgi apparatus, also known as the Golgi complex or Golgi body, is an organelle found in most eukaryotic cells.

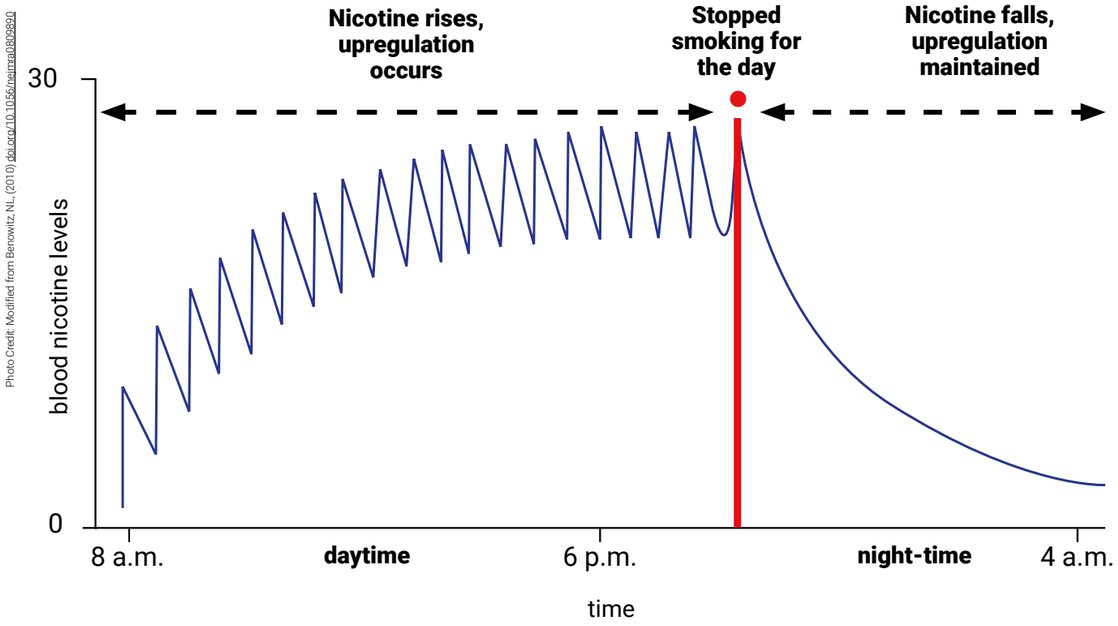


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Figure 1. Intermittent exposure to nicotine during the day results in overall plasma levels to rise and plateau until smoking stops and nicotine levels rapidly decline. Each cigarette causes rapid spiking of nicotine in the blood.

feelings of withdrawal. What causes the upregulation of these nicotinic receptors, and what are the underlying mechanisms at play? An understanding of the mechanisms of both nicotine addiction and smoking cessation drugs is needed for the design of new treatments.

Golgi satellites – the key organelles in nicotine addiction

The Golgi organelles of cells are 'protein packaging factories' that package proteins and lipids into vesicles that then travel to the cell membrane. The central Golgi apparatus forms part of a cell's secretory pathway, an essential process that enables protein release from cells that is vital for normal cell function. Processing of proteins in this 'factory' converts the proteins' sugars into more complex forms that expands their functionality. One such complex glycosylation is called sialylation, where a carbohydrate unit, sialic acid, is added to the other glycans attached to the protein.

The research team recently discovered that when neurons are stimulated, the Golgi apparatus appears to disperse into hundreds of smaller Golgi satellites. This occurs during high neuronal electrical activity as well as during exposure to nicotine. These miniature Golgi satellites emerge from another part of the secretory pathway, the endoplasmic reticulum, and associate nearby at membrane domains, especially synapses. There, they provide a glycosylation service for local proteins in neurons. Included in the proteins glycosylated at these stations are α4β2Rs, which then become upregulated. This results in the addition of sialic acid to α4β2Rs, which causes the increased functional response. This α4β2R upregulation is likely central to the mechanism of nicotine addiction. More broadly, this discovery sheds light on how proteins made and transported to synapses are altered. These findings have implications for understanding memory formation and neurological diseases, as well as addiction.

> **The discovery of the role of Golgi satellites in nicotine addiction and as a mechanism of smoking cessation drugs could be pivotal in developing new drugs.**

Kicking the habit

Smoking cessation treatments include nicotine replacement therapy, commonly known as 'patches' and varenicline, commercially sold as Chantix, a prescribed drug with the highest smoking cessation success rates of about 50%. Varenicline is widely used, although the UK and Europe have recently withdrawn it from the market. Both nicotine and varenicline have an affinity for α4β2Rs, meaning they bind to and interact with these receptors. Despite the success of varenicline, we do not know how the drug works in smoking cessation.

Varenicline's immediate anti-nicotine effects are well understood, but in their most recent paper, the team have uncovered its long-term effects. In a breakthrough study, Green and colleagues used in vitro and in vivo imaging of mice brains to show how varenicline gets trapped inside acidic vesicles that also contain α4β2Rs. Nicotine is less acidic, and although it accumulates in vesicles, it does not get trapped, resulting in a brief lifetime in the brain of 1-2 hours. Ligand trapping of varenicline occurs because of its high acidity and affinity for α4β2Rs. In fact, fluorescent imaging shows that this trapping occurs in the Golgi satellites. The findings show that varenicline is slowly released over time and well after nicotine has dissipated after smoking. Varenicline is most effective when released as smoking ends and targets the functionally upregulated α4β2Rs.

This research significantly expands our knowledge of nicotine addiction. It could help us understand addiction to other drugs, such as opioids or cocaine, due to similar chemical properties that may make them susceptible to being trapped in vesicles. The discovery of the role of Golgi satellites in nicotine addiction and as a mechanism of smoking cessation drugs could be pivotal in developing new drugs. The researchers maintain that further studies will expand our knowledge of nicotine addiction and aid in the design, development, and testing of smoking cessation drugs. More effective therapies would help people quit smoking and prevent needless deaths.

Personal response

Are there any things that have surprised you during your studies on this topic?

What has surprised me most is how little is known about the cell biology of neurons and glia in the brain. Golgi satellites appear to be the cell biological adaptations that neurons and glia have made to adjust to their unusual morphological features and size. Our lab and others have just begun to characterise where Golgi satellites are found in neurons and their different functions. For example, the rapid changes in protein glycosylation that have recently been shown to occur within seconds of stimulations at synapses may be a mechanism triggering synaptic plasticity (Boll et al, 2020). Given the long distance of the Golgi apparatus from most synapses, these changes cannot be mediated at the normal site of glycosylation, which is the Golgi apparatus found in the soma of neurons. Instead, neurons have adapted to the need of synapses to rapidly change the glycosylation of proteins at synapses by positioning Golgi satellites at synapses. Despite their abundance in active neurons, Golgi satellites had been missed because the usual Golgi apparatus identifiers are not found in Golgi satellites. Thus, it is possible that other adaptations have also been overlooked in neurons and glia.

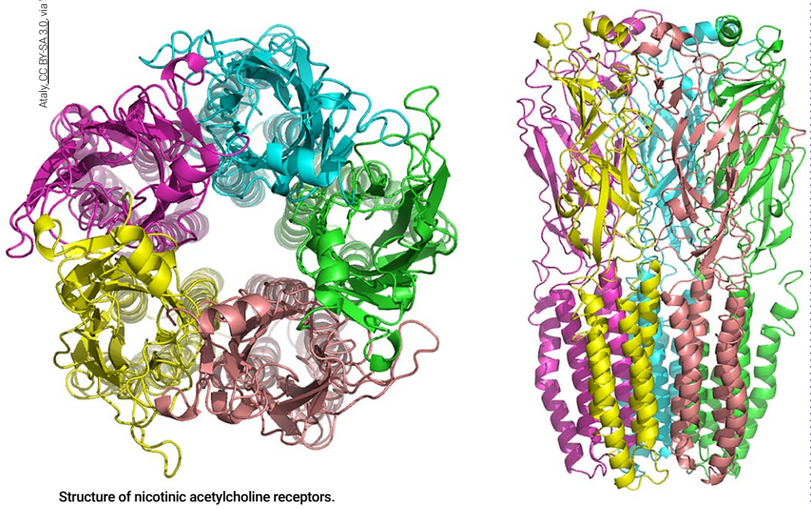
Further, our lab and others have observed that, with increased activity, the Golgi apparatus appears to disperse into many smaller 'fragments', a process called 'Golgi fragmentation'. Golgi fragmentation is a feature of several neurodegenerative disorders and assumed to be deleterious, resulting in dysregulated protein glycosylation. However, it is possible that Golgi fragmentation is not pathological but instead adaptive, a homeostatic response by the Golgi to changes in neuronal excitability. This possibility needs to be further explored, especially with respect to neurodegenerative disorders.

What should be the next step to take this research forward in terms of developing new smoking cessation drugs?

The next steps towards developing new drugs would be to understand the molecular features of varenicline that mediate its trapping in Golgi satellites and its release. If these drug features are identified, their modification could lead to new compounds that improve the effectiveness of varenicline as a smoking cessation drug and/or to reduce its side effects. Another area to investigate are the changes that occur when varenicline interacts with the functionally upregulated α4β2Rs after smoking ends. Specifically, how do the drug interactions affect the state of the upregulated receptors?

How could this work be extended and applied in the study of other addictions?

All so-called drugs of abuse, except ethanol and THC, have chemical properties like nicotine and varenicline that cause them to be either concentrated or trapped in Golgi satellites. This raises questions about the proteins or 'receptors' to which the drugs bind to, unlike nicotine-like ligands. These other receptors are all membrane proteins that traffic through Golgi membranes for their processing in the secretory pathway. Do these receptors, like α4β2Rs, traffic through Golgi satellites at synapses where they undergo glycosylation changes?



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Structure of nicotinic acetylcholine receptors.

Details

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Bio

William 'Bill' Green, PhD, is a professor at the Department of Neurobiology, University of Chicago and a Whitman Investigator at the Marine Biological Laboratory in Woods Hole, Massachusetts. He has degrees from the University of Toronto and Cornell University. He researches the cell biology of neurons and how synapses form and change with activity and disease. One of his main interests is to understand nicotine addiction and how to treat it.

Further reading

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