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Perspective
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The Birth of Antihypertensive Therapy

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Abstract

Prior to the groundbreaking studies of a few pharmacologists, the treatment of hypertension had loomed a formidable obstacle to physicians. The discovery of new directions in anti-hypertensive therapy created a paradigm for drug action at specific cellular sites. This review recounts the early work that led to the successful treatment of this common malady. Hexamethonium represented a milestone in the treatment of hypertension when it was developed by William Paton and Eleanor Zaimis. James Black, in his discovery of propranolol, made one of the most important contributions to clinical medicine and pharmacology in the twentieth century. Albrecht Fleckenstein's finding that calcium antagonist's block excitation-contraction coupling represented another major advance in the pharmacotherapy of anti-hypertensive agents. The common strategy speaks to achieving selectivity of drug action by applying basic physiological and pharmacological principles to the actions of various compounds on specific sites or receptors. The implications of these diverse discoveries were far reaching, spurred new directions in anti-hypertensive therapy, and created a paradigm for drug action at specific cellular sites. The work underscores the remarkable advances made in treating hypertension and other cardiovascular diseases when key factors in regulating blood pressure were recognized and exploited.

Keywords: Eleanor Zaimis; Albrecht Fleckenstein; Methonium; Nicotinic receptors; Calcium channels; Anti-hypertensive therapy

Objectives

Essential hypertension is one of the most common human disorders. This article describes the early events that provided knowledge about the basic physiological principles that govern blood pressure, which ultimately led to the proliferation of medications that control this malady. Drug discovery is a step-wise process that is often built upon the prior accumulation of knowledge in the field of physiology. There now exists a large number of drugs that can control blood pressure by diverse mechanisms; however, it is not my intention to provide basic information about antihypertensive drugs that can be readily found in textbooks of pharmacology, but to offer perspective as to how gifted researchers spawned the development of these drugs by forging key conceptual advances.

Ganglionic Blocking Agents

Sir William Paton (1917-1993) and Eleanor Zaimis (1915-1982).

The role of methonium compounds in the development of antihypertensive therapy actually began in the late 1850's when Claude Bernard showed that the site of action of curare was the neuromuscular junction (Figure 1) [1]. Seven decades later, Henry Dale identified two main classes of receptors for acetylcholine (muscarinic and nicotinic) (Figure 1). However, it was not until 1935 that the chemical relationship between the cholinergic neurotransmitter acetylcholine and curare was established when Harold King isolated d-tubocurarine from crude curare. He further demonstrated that its chemical structure resembled acetylcholine by consisting of a quaterna nitrogen atom in the amine group [2]. Although William Paton, working at the Medical Research Institute in London in the late 1940's, collaborated with a number of talented colleagues on the study of methonium compounds, attention had been focused on the ability of these compounds to elicit histamine release. At the time, Paton was one of the most accomplished, insightful, and effective leaders in the biomedical sciences (Figure 2). His achievements would be recognized at an early age, when he became Professor of Pharmacology at the University of Oxford. Paton was later named a Fellow of the Royal Society in 1956 and knighted in 1979 [3].

Eleanor Zaimis (Figure 3) played a most vital role in enhancing Paton's reputation when she moved to the National Institute for Medical Research in 1949. Although she worked in Paton's

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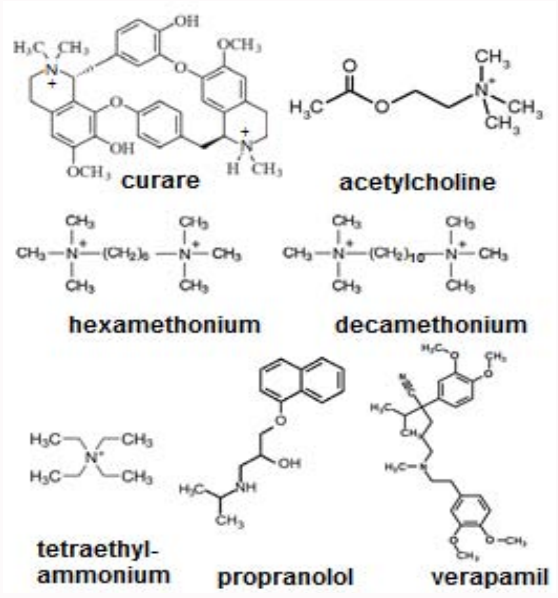

Figure 1: Compounds instrumental in the development of antihypertensive agents.

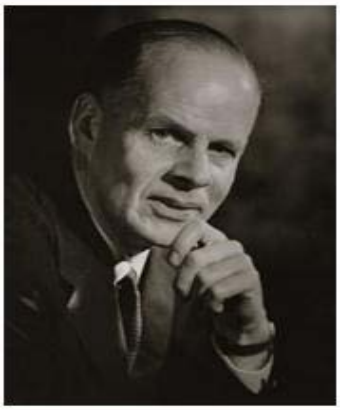

Figure 2: Sir William D.M. Paton. (Courtesy of British Pharmacological Society; Pharmacology Hall of Fame. 19-Nov.2015. https://www.bps.ac.uk/about-pharmacology/pharmacology-hall-of-fame/articles/sir-william-paton).

laboratory for only a short time, Zaimis made the most of the opportunity. There, she exploited her expertise in structure-activity relationships to carry out her groundbreaking collaboration with William Paton on ammonium compounds. They reasoned that since curare was an alkaloid containing two quaternary nitrogen groups, its paralytic activity might be due to the presence of two such cation groups at some optimal distance apart. After synthesizing a series of dibasic compounds with methylene groups from C_2 to C_{12} [4], Zaimis carried out a pharmacological analysis of these substances. She demonstrated that the ability of acetylcholine to elicit skeletal muscle contraction could be distinguished by hexamethonium (C_6) and decamethonium (C_{10}), two compounds that differed only in the number of methylene groups between two quaternary nitrogen atoms (Figure 1).

After publishing a short note to Nature in 1948 [5] describing the curare-like action of decamethonium, later that year Paton and Zaimis published another article in the same journal on the clinical potentialities of certain bisquaternary bases causing neuromuscular

Figure 3: Eleanor Zaimis.(Courtesy of the British Pharmacological Society. Published 19 Nov. 2015 in Pharmacology Hall of Fame. https//www.bps.ac.uk/about/who-we-are).

and ganglionic blockade [6]. Their findings that hexamethonium displayed a peak in potency for ganglionic block and decamethonium showed a peak in potency for neuromuscular block provided the first piece of evidence that the nicotinic cholinergic receptors at autonomic ganglia and their associated ion channels at the neuromuscular junction were distinct entities. These studies spawned future work on sympathetic ganglia as a site of action of anti-hypertensive agents (see below).In addition, a purified extract of curare was introduced into general anesthesia as a muscle relaxant by Harold Griffith in 1942 [7], which was eventually replaced by decamethonium, and then by suxamethonium. It is of interest to note that during this time Barlow and Ing at Oxford had also been investigating the curare-like action of a number of bisquaternary ammonium salts in which the nitrogen atoms were separated by methylene groups of various lengths. Therefore, the initial publication by Paton and Zaimis in Nature appeared immediately following that of Barlow and Ing [8]. At the time of the discovery of hexamethonium, there was no effective treatment for severe hypertension, and Draconian measures such as bilateral adrenalectomy and sympathectomy were employed to limit the symptoms of "malignant" hypertension [9]. The introduction of ganglionic blocking drugs for the treatment of severe hypertension began the modern era of pharmacotherapy of this prevalent disease. Joshua Burn and Sir Henry Dale first described the "nicotine-depolarizing" action of tetraethylammonium (TEA) on autonomic ganglia in 1915 [10]; but this observation was overlooked until George Acheson and co-workers described its action on ganglia and the cardiovascular system [11]. Although Acheson proposed the use of TEA in the treatment of hypertension its hypotensive effect was never considered clinically significant because of its short duration of action (Figure 1).The controversy as to whether hypertension was a primary disease or only a symptom of an underlying cause further compromised effective treatment, which at the time was limited to bed rest, sedation, and vene section. Many of the patients afflicted with this disease were left untreated.

At the beginning of their investigations, Paton and Zaimis were interested in the ammonium compounds solely as research tools; and only in their second paper in Nature did they suggest that hexamethonium might be useful in the treatment of hypertension and vascular disease [12]. Nevertheless, when the Medical Research Council organized a committee chaired by Paton to investigate the possible clinical effects of the methonium compounds, Paton and Zaimis courageously tested several of these compounds on themselves

Figure 4: Sir James Black.(Taken from Sir James W. Black-Biographical. The Nobel Prize in Physiology or Medicine 1988. Nobel Foundation; Stockholm, 1989). https://www.nobelprize.org/nobel_prizes/medicine/laureates/1988/black-bio.html.

Figure 5: Albrecht Fleckenstein. Recipient of Einstein World of Science Award, 1991world Cultural Council. https:en.wikipedia.org/wiki/Albert_einstein_World_Award_of_Science.

with the assistance of an anesthesiologist.

Although hexamethonium was described by Paton as specific both in its structure and action [13], it was much less than an ideal drug. By causing a non-depolarizing blockade of both sympathetic and parasympathetic ganglia, hexamethonium produced what was characterized as a "medical sympathectomy", or somewhat sardonically as the "hexamethonium man". Side effects included postural hypotension, constipation, blurred vision, and fainting. Another drawback of the quaternary drug was that it had to be given subcutaneously. Despite its limitations, hexamethonium represented a milestone in the treatment of hypertension. In 1951 a Lancet editorial stated that "Early reports leave no doubt that the methonium compounds are the most powerful hypotensive agents yet developed". The idea that hypertension could be ameliorated by pharmacotherapy had taken root [14]. However, the poorly absorbed hexamethonium was soon preempted by mecamylamine and still later by adrenergic receptor blockers such as propranolol (Figure 1; see below), which circumvented parasympathetic side effects. Although ganglionic blockade is no longer employed to reduce systemic blood pressure, the introduction of even more useful and specific antihypertensive agents, such as diuretics, calcium channel blockers and angiotensin-converting enzyme inhibitors, now offer a diverse number of medications to treat this disease.

At the end of 1948, Zaimis left the Institute for the School of Pharmacy at the University of London, where she continued to co-author publications with Paton until 1952. She was appointed Head of the Department of Pharmacology at the Royal Free Hospital School of Medicine in 1954 at the age of 39; she served in that position at the women's medical school for the next 25 years [15]

Beta Adrenergic Blocking Agents

Sir James Black (1924-2010)

The studies of Paton and Zaimis created a paradigm for drug action by demonstrating that chemically related drugs act at two types of nicotinic receptors, one in autonomic ganglia, the other in skeletal muscle. However, the non-selectivity of action of agents that blocked autonomic ganglia obviously mandated for new and more selective antihypertensive agents. James Black (Figure 4) became involved in drug research and a different mode of therapy during the 1950's at Glasgow University Veterinary School, when he developed interest in the relation between epinephrine and cardiovascular disease. Because he believed that the advent of better anti-hypertensive medication could not be realized by employing existing methodology, Black formulated a plan to develop drugs that would block the effects of epinephrine by decreasing the demand for oxygen, rather than increasing it [16]. A move to the Imperial Chemical Industries (ICI) Pharmaceutical Division in the late 1950'sprovided the impetus to embark on this project.

Black realized that it was crucial to understand the physiological factors that regulated blood pressure in order to recognize the key elements in altering the process. Black had become aware of the delineation by Raymond Ahlquist in 1948 of adrenergic receptors into alpha and beta components, which caused different tissue responses. In this new classification, the anti-epinephine drugs were alpha antagonists, and isoproterenol was a stimulant of beta-receptors [17]. This concept provided Black with the theoretical basis for developing beta-adrenergic receptor blocking agents which were derivatives of isoproterenol. However, at the time only a small group of scientists were able to describe by mathematical equations the binding of two mutually exclusive compounds at the same population of binding sites. For example, the Schild plot defined the properties of competitive antagonism by analyzing the binding of a competitive antagonist to its receptor [18].

By the 1950's, alpha-adrenergic receptor blocking agents were a well-recognized class of pharmacological agents which reversed the rise in blood pressure produced by epinephrine, but did not affect the accompanying tachycardia. Black argued that the imbalance between the oxygen supply to the heart and the demand was quite small to the extent that reducing the work of the heart would be a useful approach to developing a beta-receptor blocking drug [19]. This new approach to the problem formulated by James Black provided impetus to an area of research that was previously lacking. Thus, by judiciously utilizing the analytical power of competitive antagonists, Black was successful in developing drugs that lowered blood pressure by limiting the oxygen demands of the heart. As a result, Black would later become known as the *Father of Analytical Pharmacology* [20]. When the project of beta-receptor blockade was instituted by Black in 1956, no useful antagonists were available. However, in 1957, Powell and Slater at Eli Lilly synthesized dichloro-isoproterenol, an agent that blocked beta-adrenoceptors [21]. In 1958, Black and colleagues at Imperial Chemical Industries found that this compound lacked agonist activity, although it was later determined to be a partial agonist. The potential for clinical utility of this compound was short-lived because of its low potency and its stimulatory properties [22]. Black continued to develop analogs sufficiently similar to epinephrine and isoproterenol to block adrenergic receptors, but different enough not to cause their activation. Chemical modification of these compounds

by Black and Stephenson eventually yielded pronethalol (ICI 38-174) [23]. This analog expressed antagonist activity without significant agonist activity in both ventricular and a trial tissue. Pronethalol also reduced the resting heart rate and depressed increments from stimulation of the sympathetic nerve. However, its clinical use to treat cardiac arrhythmias was brought to a halt by its production of malignant tumors in mice. Undaunted, Black then introduced the more active analog propranolol (Figure 1), which was hailed as the greatest advance in the treatment of cardiovascular disorders since digitalis in the 18th century. By blocking the cardiovascular response of adrenergic receptor agonists, propranolol was much more active than pronethalol, but was devoid of sympathomimetic activity [24]. By introducing propranolol, Black was responsible for producing cell receptor modulators of cardiovascular disorders, which not only led to the reduction of blood pressure in hyper tensives and the chest pain of angina pectoris, but he spawned the explosive developments that were to occur in cardiovascular pharmacology in the ensuing years. The advent of propranolol also initiated basic studies of the physiological role of beta-receptors, which were subsequently divided into two classes, beta-1 and beta-2. As a result, Black's work fostered a much deeper understanding of the basic pharmacology of the autonomic nervous system that existed at this time. Then, in 1964 at Smith-Kline and French, Black's unique approach to drug discovery led him to develop the histamine antagonist cimetidine, which unequivocally brought about the emergence of the concept of *drug receptors* into scientific lore [25]. Because Black divided his successful career between academia and industry, he was endowed with the broad experience that facilitated his work on drug discovery. Perceiving a link between clinical medicine and academic pharmacology, he advocated an approach to a problem that emanated from the laboratory, rather than from the clinic. In addition, Black always stressed that scientists should focus on the most significant and interesting questions, regardless of the financial opportunities that might accrue from the work. Black's discoveries are considered to be among the most important contributions to clinincal medicine and the discipline of pharmacology in the twentieth century. For his work on drug antagonism, Sir James Black was awarded the Nobel Prize in Medicine in 1988, together with Gertrude Elion and George Hitchings, for their discoveries of important principles for drug treatment [26]. In addition, the award of the Nobel Prize to James Black was a milestone in that it heralded the contribution made by industrial scientists to the discipline of pharmacology [27].

Calcium Antagonists

Albrecht Fleckenstein (1917-1992).

The evolution of hypertensive therapy continued with major advances in cardiovascular physiology/pharmacology and electrophysiology, which dealt with the importance of calcium in maintaining cardiac contraction. This concept was first reported by Sydney Ringer in the 1880's [28] and has been followed by a massive amount of evidence that calcium ions are required during excitation (depolarization) to activate the biochemical processes that utilize ATP for mechanical contractility [29]. Albrecht Fleckenstein (Figure 5), Professor of Physiology and Head of the Physiology Institute at the University of Freiburg, Germany, was brought into this arena by the scientists at the German company Bayer Pharmaceuticals, when they sent verapamil to him for testing in 1964 [30]. Fleckenstein reported that verapamil mimicked the effects of simple withdrawal of calcium by blocking the contractile force and the oxygen requirement of cardiac muscle, without affecting the sodium-dependent action potential [31]. Further studies by Fleckenstein and colleagues demonstrated that calcium antagonists interfered with uptake of calcium into the myocardium and excitation-contraction coupling in vascular smooth muscle [32]. These actions, which were reversed by the addition high concentrations of calcium, explained the effects of these drugs as vasodilators, and as cardiac depressants. Verapamil was the first calcium antagonist to be introduced into pharmacotherapy in many countries in 1967 (Figure 1) [33]. The drug's potential for reducing peripheral vascular resistance led to studies on patients with hypertension and its approval for general use by the Federal Drug Administration in the United States in 1982 [34]. The study of antihypertensive therapy was greatly enhanced by new techniques. The advent of single channel recording by patch clamp provided new technologies for observing the kinetic properties of individual ion channels in the cell membrane [35]. The properties of these channels can be modulated by a change in the membrane potential or binding of an agonist to a receptor. The voltage clamp technique not only enabled measurements of both calcium and sodium currents, but the major calcium currents could be distinguished by single channel conductance, voltage-dependent inactivation, regulation by cAMP and specific inhibition by calcium antagonist drugs [36]. Such findings revealed that calcium antagonists not only block the inhibition of calcium-dependent contractility, but they suppress calcium-dependent membrane excitation and the resulting activation of ionic currents. Albrecht Fleckenstein's discovery that the calcium antagonists block excitation-contraction coupling represented another major advance in the pharmacotherapy of anti-hypertensive agents. They are effective in treating patients with hypertension by decreasing cytosolic calcium concentrations and indirectly by activating calcium-dependent calcium release by ryanodine-sensitive release channels in the sarcoplasmic reticulum. Fleckenstein coined the term *calcium antagonist* to define this new group of agents that interfere with the uptake of calcium into the myocardium, and he became known as the 'father of calcium antagonism' [37]. Shortly after verapamil became a key component in the treatment of hypertension, ischemic heart disease and certain cardiac arrhythmias, nifedipine was discovered in 1969, developed by Bayer in the early 1970's (BAYa1040), [38] and approved for use in the United States in 1982 [39]. Nifedipine displays selectivity for smooth muscle, while verapamil shows less selectivity in this regard. A number of other calcium antagonists are now available, including diltiazem, amlodipine, felodipine, and nicardipine [40]. As a heterogeneous group of compounds with a specific mechanism of action, they have not only provided invaluable tools in elucidating the key roles of calcium in a variety of biological systems, these drugs have extended the therapeutic landscape for the treatment of hypertension, as well as other cardiovascular disorders [41].

Perspectives

This article has attempted to show how our concepts of hypertensive therapy have evolved over the years. The resonating theme intrinsic to the strategy employed by the researchers cited in this article speaks to achieving selectivity of drug action by applying basic physiological and pharmacological principles to the actions of various compounds on specific sites or receptors. Prior to the groundbreaking studies of the talented scientists described herein, the treatment of hypertension had loomed a formidable obstacle to physicians. This article reaffirms the remarkable progress that was made during the last half century in treating hypertension and other cardiovascular diseases after key factors in regulating blood pressure

were recognized and exploited.

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