A Brief History of Natural Products Chemistry

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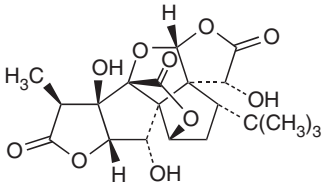
To give an account of the rich history of natural products chemistry in a short essay is a daunting task. This brief outline begins with a description of ancient folk medicine, and continues with an outline of some of the major conceptual and experimental advances that have been made from the early 19th century through about 1960, the start of the modern era of natural products chemistry. Achievements of living chemists are noted only minimally, usually in the context of related topics within the text. More recent developments are reviewed within the individual chapters of the present volumes, written by experts in each field. The subheading follow, in part, the sequence of topics presented in Volumes 1-8.

Ethnobotany and "natural products chemistry."

Except for minerals and synthetic materials, our surroundings consist entirely of organic natural products, either of prebiotic organic origins or from microbial, plant, or animal sources. These materials include polyketides, terpenoids, amino acids, proteins, carbohydrates, lipids, nucleic acid bases, RNA and DNA, etc. Natural products chemistry can be thought of as originating from mankind's curiosity about odor, taste, color, and cures for diseases. Folk interest in treatments for pain, for food-poisoning and other maladies, and in hallucinogens appears to go back to the dawn of humanity

For centuries, China has led the world in the use of natural products for healing. One of the earliest health science anthologies in China is the Nei Ching, whose authorship is attributed to the legendary Yellow Emperor (30th century BC), although it is said that the dates were backdated from the 3rd century by compilers. Excavation of a Han Dynasty (206 BC-220 AD 220) tomb in Hunan Province in 1974 unearthed decayed books, written on silk, bamboo and wood, which filled a critical gap between the dawn of medicine up to the classic Nei Ching; book 5 of these excavated documents lists 151 medical materials of plant origin. Generally regarded as the oldest compilation of Chinese herbs is Shen Nung Pen Ts'ao Ching (Catalog of Herbs by Shen Nung), which is believed to have been revised during the Han Dynasty; it lists 365 materials. Numerous revisions and enlargements of Pen Ts'ao (herb) were undertaken by physicians in subsequent dynasties, the ultimate being the Pen Ts'ao Kang Mu (General Catalog of Herbs) written by Li Shih-Chen over a period of 27 years during the Ming Dynasty (1573-1620), which records 1898 herbal drugs and 8,160 prescriptions. This was circulated in Japan around 1620 and translated, and has made a huge impact on subsequent herbal studies in Japan; however, it has not been translated into English. The number of medicinal herbs used in 1979 in China numbered 5,267. One of the most famous of the Chinese folk herbs is the ginseng root *Panax ginseng*, used for health maintenance and treatment of various diseases. The active principles were thought to be the saponins called ginsenosides but this is now doubtful; the effects could well be synergistic between saponins, flavonoids, etc. Another popular folk drug, the extract of the Ginkgo tree, *Ginkgo biloba* L., the only surviving species of the Paleozoic era (250 million years ago) family which became extinct during the last few million years, is mentioned in the Chinese Materia Medica to have an effect in improving memory and sharpening mental alertness. The main constituents responsible for this are now understood to be ginkgolides **1** and flavonoids, but again not

much else is known. Clarifying the active constituents and mode of (synergistic) bioactivity of Chinese herbs is a challenging task that has yet to be fully addressed.

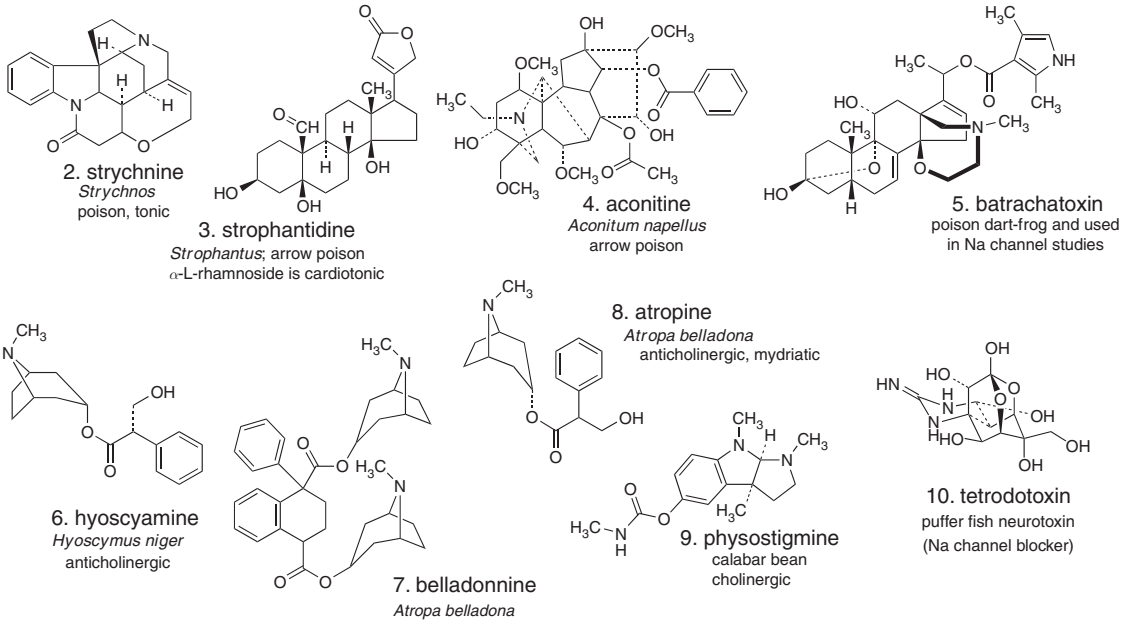


1. ginkgolide B
Ginkgo biloba
PAF (platelet activating factor)

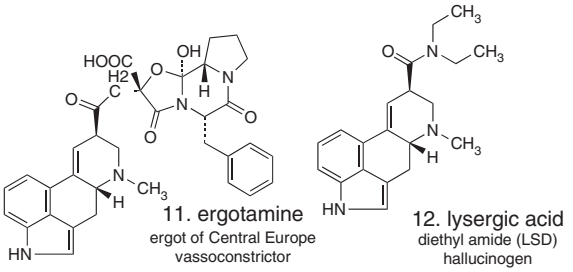
The Assyrians left 660 clay tablets describing 1000 medicinal plants used around 1900-400 BC, but the best insight into ancient pharmacy is provided by the two scripts left by the ancient Egyptians who were masters of human anatomy and surgery because of their extensive mummification practices. The Edwin Smith Surgical Papyrus purchased by Smith in 1862 in Luxor (now in the New York Academy of Sciences collection), is one of the most important medicinal documents of the ancient Nile Valley, and describes the healer's involvement in surgery, prescription and healing practices using plants, animals and minerals. The Ebers Papyrus, also purchased by Edwin Smith in 1862, and then acquired by Egyptologist George Ebers in 1872, describes 800 remedies using plants, animals, minerals and magic. Indian medicine also has a long history, possibly dating back to the 2nd millennium BC. The Indian materia medica consisted mainly of vegetable drugs prepared from plants but also used animals, bones, and minerals, e.g., sulfur, arsenic, lead, copper sulfate, and gold. Ancient Greece inherited much from Egypt, India and China, and underwent a gradual transition from magic to science. Pythagoras (580-500 BC) influenced the medical thinkers of his time, including Aristotle (384-322 BC), who in turn affected the medical practices of another influential Greek physician Galen (129-216). The Iranian physician Avicenna (980-1037) is noted for his contributions to Aristotelian philosophy and medicine, while the German-Swiss physician and alchemist Paracelsus (1493-1541) was an early champion who established the role of chemistry in medicine.

The rain-forests in Central and South America and Africa are known to be particularly abundant in various organisms of interest to our lives because of their rich biodiversity, intense competition, and the necessity for self-defense. However, since folk-treatments are transmitted verbally to the next generation via shamans who naturally have a tendency to keep their plant and animal sources confidential, the recipes tend to get lost, particularly with deforestation of rain forests and the encroachment of "civilization." Studies on folk medicine, hallucinogens and shamanism of the Central and South American Indians conducted by Richard Schultes (Harvard Botanical Museum, emeritus) have led to renewed recent activity by ethnobotanists, recording the knowledge of shamans, assembling herbaria, and transmitting the record learning to the village.

Extracts of toxic plants and animals have been used throughout the world for thousands of years for hunting and murder. These include the various arrow poisons used all over the world. Strychnos and Chondrodendron (containing strychnine **2**, etc.) was used in South America and called "curare", *Strophanthus* (strophantidine **3**, etc.) was used in Africa, the latex of the upas tree Antiaris toxicaria (cardiac glycosides) was used in Java, while Aconitum napellus, which appears in Greek mythology (aconitine **4**) was used in medieval Europe and Hokkaido (by the Ainus). The Colombian arrow poison is from frogs (batrachotoxins **5**; 200 toxins have been isolated from frogs by B. Witkop, J. Daly at NIH). Extracts of *Hyoscyamus niger* and *Atropa belladonna* contain the toxic tropane alkaloids, e.g., hyoscyamine **6**, belladonnine **7** and atropine **8**. The belladonna berry juice (atropine) which dilates the eye pupils was used during the Renaissance by ladies to produce doe-like eyes (belladona means beautiful woman). The Efik people in Calabar, southeastern Nigeria, used extracts of the calabar bean known as esere (physostigmine **9**) for unmasking witches. The ancient Egyptians and Chinese knew of the toxic effect of the puffer fish, fugu, which contains the neurotoxin tetrodotoxin **10** (Y. Hirata, K. Tsuda, R.B. Woodward).

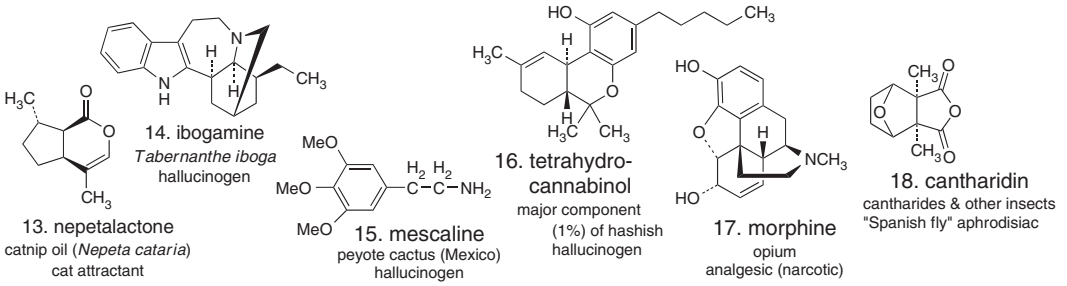


When rye is infected by the fungus *Claviceps purpurea*, the toxin ergotamine **11** and a number of ergot alkaloids are produced. These cause ergotism or the "devil's curse", "St. Anthony's fire," which leads to convulsions, miscarriages, loss of arms and legs, dry gangrene and death. Epidemics of ergotism occurred in medieval times in villages throughout Europe, killing tens of thousands of people and livestock; Julius Caesar's legions were destroyed by ergotism during a campaign in Gaul, while in 994 AD an estimated 50,000 people died in an epidemic in France. As recently as in 1926, a total of 11,000 cases of ergotism were reported in a region close to the Urals. It has been suggested that the witch hysteria that occurred in Salem, Massachusetts, might have been due to a mild outbreak of ergotism. Lysergic acid diethylamide (LSD) **12** was first prepared by A. Hofmann, Sandoz Laboratories, Basel, in 1943 during efforts to improve the physiological effects of the ergot alkaloids when he accidentally inhaled it. "On Friday afternoon, April 16, 1943, he wrote "I was seized by a sensation of restlessness. . ." He went home from the laboratory and "perceived an uninterrupted stream of fantastic dreams, . . ." (*Helv. Chim. Acta*).

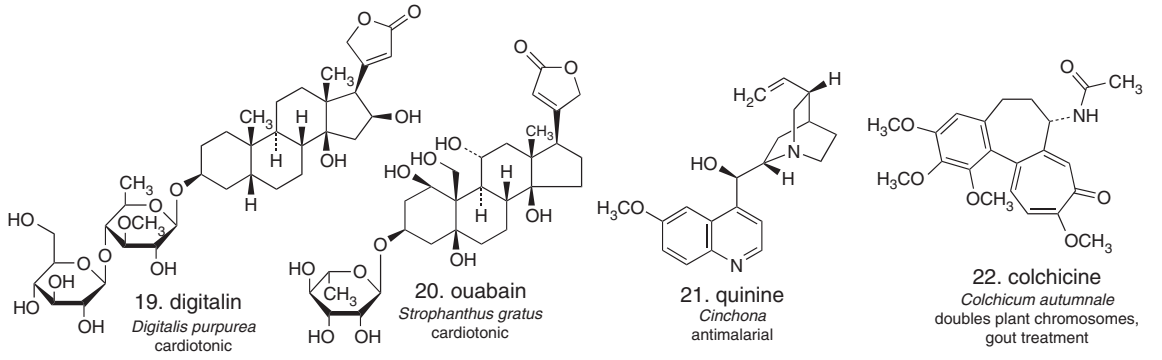


Numerous psychedelic plants have been used since ancient times, producing visions, mystical fantasies (cats and tigers also seem to have fantasies, see nepetalactone **13** below), sensations of flying, glorious feelings in warriors before battle, etc. The ethnobotanists Wasson and Schultes identified "ololiqui", an important Aztec concoction, as the seeds of the morning glory *Rivea corymbosa* and gave the seeds to Hofmann who found that they contained lysergic acid amides similar but less potent than LSD. Iboga, a powerful hallucinogen from the root of the African shrub *Tabernanthe iboga*, is used by the Bwiti cult in Central Africa who chew the roots to obtain relief from fatigue and hunger; it contains the alkaloid ibogamine **14**. The powerful hallucinogen used for thousands of years by the American Indians, the peyote cactus, contains mescaline **15** and other alkaloids. The Indian hemp plant, *Cannabis sativa*, has been used for making rope since 3000 BC, but when it is used for its

pleasure-giving effects it is called cannabis and has been known in central Asia, China, India and the Near East since ancient times. Marijuana, hashish (named after the Persian founder of the Assassins of the 11th century, Hasan-e Sabbah), charas, ghanja, bhang, kef, and dagga are names given to various preparations of the hemp plant. The constituent responsible for the mind-altering effect is Δ^1-tetrahydrocannabinol **16** (also called Δ^9-THC), the content in the plant being 1%. R. Mechoulam (1930-, Hebrew U.) has been the principal worker in the cannabinoids, including structure determination and synthesis of Δ^9-THC (1964-present); the Israeli police have also made a contribution by providing Mechoulam with a constant supply of marijuana. Opium (morphine **17**) is another ancient drug used for a variety of pain-relieves and it is documented that the Sumerians used poppy as early as 4000 BC; the narcotic effect is present only in seeds before they are fully formed. The irritating secretions of the blister beetle, e.g., *Mylabris* and the European species *Lytta vesicatoria*, commonly called Spanish fly was used medically as a topical skin irritant to remove warts but was also a major ingredient in so-called love potions (constituent is cantharidin **18**, stereospecific synthesis in 1951, G. Stork, 1921-; prep. scale high-pressure Diels-Alder synthesis in 1985, W.G. Dauben, 1919-96).

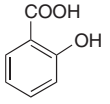


Plants have been used for centuries for treatment of heart ailment, the most important being the foxgloves *Digitalis purpurea* and *D. lanata* (digitalin **19**, diginin) and *Strophanthus gratus* (ouabain **20**). The bark of cinchona *Cinchona officinalis* (called quina-quina by the Indians) has been used widely among the Indians in the Andes against malaria, which is still one of the major infectious diseases; its most important alkaloid is quinine. The British protected themselves against malaria during the occupation of India through gin and tonic (quinine **21**)(!). The stimulant coca, used by the Incas around the 10th century, was introduced into Europe by the conquistadors; coca beans are also commonly chewed in West Africa. Wine making was already practiced in the Middle East 6,000-8,000 years ago; Moors made date wines, the Japanese rice wine, the Vikings honey mead, the Incas maize chicha. It is said that the Babylonians made beer using yeast 5,000-6,000 years ago. As shown above in parentheses, alkaloids are the major constituents of the herbal plants and extracts used for centuries, but it was not until the early 19th century that the active principles were isolated in pure form, e.g., morphine **17** (1816), strychnine **2** (1817), atropine **8** (1819), quinine **21** (1820), and colchicine **22** (1820). It was a century later when the structures of these compounds were finally elucidated.



Dawn of organic chemistry, early structural studies, modern methodology

The term "organic compound" to define compounds made by and isolated from living organisms was coined in 1807 by the Swedish chemist Jons Jacob Berzelius (1779-1848), a founder of today's chemistry, who developed the modern system of symbols and formulas in chemistry, made a remarkably accurate table of atomic weights and analyzed many chemicals. At that time it was considered that organic compounds could not be synthesized from inorganic materials, *in vitro*. However, Friedrich Wöhler (1800-1882), a medical doctor from Heidelberg who was starting his chemical career at a technical school in Berlin, attempted in 1828 to make "ammonium cyanate", which had been assigned a wrong structure, by heating the two inorganic salts potassium cyanate and ammonium sulfate; this led to the unexpected isolation of white crystals which were identical to the urea from urine, a typical organic compound. This well-known incident marked the beginning of organic chemistry. With the preparation of acetic acid from inorganic material in 1845 by Hermann Kolbe (1818-1884), Leipzig, the myth surrounding organic compounds, in which they were associated with some vitalism was brought to an end and organic chemistry became the chemistry of carbon compounds. The same Kolbe was involved in the development of aspirin, one of the earliest and most important success stories in natural products chemistry. Salicylic acid **23** from the leaf of wintergreen plant had long been used as a pain reliever, especially in treating arthritis and gout. The inexpensive synthesis of salicylic acid from sodium phenolate and carbon dioxide by Kolbe in 1859 led to the industrial production in 1893 by the Bayer Company of acetylsalicylic acid "aspirin", still one of the most popular drugs. Aspirin is less acidic than salicylic acid and therefore causes less irritation in the mouth, throat and stomach. The remarkable mechanism of the anti-inflammatory effect of aspirin was clarified in 1974 by John Vane (1927-) who showed that it inhibits the biosynthesis of prostaglandins by irreversibly acetylating a serine residue in prostaglandin synthase. Vane shared the 1982 Nobel Prize with Bergström and Samuelsson who determined the structure of prostaglandins (see below).



23. salicylic acid

winter green leaves
keratolytic

In the early days, natural products chemistry was focused on isolating the more readily available plant and animal constituents and determining their structures. The course of structure determination in the 1940s was a complex, indirect process, combining evidence from many types of experiments. The first effort was to crystallize the unknown compound or make derivatives such as esters or 2,4-dinitrophenylhydrazones, and to repeat recrystallization until the highest and sharp melting point was reached, since prior to the advent of isolation and purification methods now taken for granted, there was no simple criterion for purity. The only chromatography was through special grade alumina (first used by M. Tswett in 1906, then re-introduced by R. Willstätter). Molecular weight estimation by the Rast method which depended on melting point depression of a sample/camphor mixture, coupled with Pregl elemental microanalysis (see below) gave the molecular formula. Functionalities such as hydroxyl, amino, and carbonyl groups were recognized on the basis of specific derivatization and crystallization, followed by re-determination of molecular formula; the change in molecular composition led to identification of the functionality. Thus, sterically hindered carbonyls, e.g., the 11-keto group of cortisone, or tertiary hydoxyls, were very difficult to pinpoint, and often had to depend on more searching experiments. Therefore, an entire paper describing the recognition of a single hydroxyl group in a complex natural product would occasionally appear in the literature. An oxygen function suggested from the molecular formula but left unaccounted for would usually be assigned to an ether.

Determination of C-methyl groups depended on Kuhn-Roth oxidation which is performed by drastic oxidation with chromic acid/sulfuric acid, reduction of excess oxidant with hydrazine, neutralization with alkali, addition of phosphoric acid, distillation of the acetic acid originating from the C-methyls, and finally its titration with alkali. However, the results were only approximate, since gem-dimethyl groups only yield one equivalent of acetic acid, while primary, secondary and tertiary methyl groups all give different yields of acetic acid. The skeletal structure of polycyclic compounds were frequently deduced on the basis of dehydrogenation reactions. It is then not surprising that the original steroid skeleton put forth by Wieland/Windaus in 1928, which depended a great deal on the production of chrysene upon Pd/C dehydrogenation, had to be revised in 1932 after several discrepancies were found (they received the Nobel prizes in 1927 and 1928 for this "extraordinarily difficult structure determination", see below).

In the following are listed some of the Nobel prizes awarded for the development of methodologies which have contributed critically to the progress in isolation protocols and structure determination. The year in which each prize was awarded is preceded by "Np".

Fritz Pregl, 1869-1930, Graz U., Np 1923. Invention of C and H micro-analysis. Improvement of Kuhlmann's microbalance enabled weighing at an accuracy of 1 microgram over a 20 g range, and refinement of carbon and hydrogen analytical methods made it possible to perform analysis with 3-4 mg of sample. His microbalance and the monograph "Quantitative organic microanalysis" (1916) profoundly influenced subsequent developments in practically all fields of chemistry and medicine.

The Svedberg, 1884-1971, Uppsala, Np 1926. Uppsala was a center for quantitative work on colloids for which the prize was awarded. His extensive study on ultracentrifugation, the first paper of which was published in the year of the award, evolved from a spring visit in 1922 to the U. of Wisconsin. The ultracentrifuge together with the electrophoresis technique developed by his student Tiselius, have profoundly influenced subsequent progress in molecular biology and biochemistry.

Arne Tiselius, 1902-1971, Ph. D. Uppsala (T. Svedberg); Uppsala, Np 1948. Assisted by a grant from the Rockefeller Foundation, Tiselius was able to use his early electrophoresis instrument to show four bands in horse blood serum, alpha, beta and gamma globulins in addition to albumin; the first paper published in 1937 brought immediate positive responses.

Archer Martin, 1910-, Ph. D. Cambridge; Med. Res. Council, Mill Hill, and Richard Synge, 1914-1994, Ph. D. Cambridge; Rowett Res. Inst., Food Res. Inst., Np 1952. They developed chromatography using two immiscible phases, gas-liquid, liquid-liquid and paper chromatography, all of which has profoundly influenced all phases of chemistry.

Frederick Sanger, 1918-, Ph.D. Cambridge (A. Neuberger); Med. Res. Council, Cambridge, Np 1958 and 1980 . His confrontation with challenging structural problems in proteins and nucleic acids led to the development of two general analytical methods, 1,2,4-fluorodinitrobenzene (DNP) for tagging free amino groups (1945) in connection with insulin sequencing studies, and the dideoxynucleotide method for sequencing DNA (1977) in connection with recombinant DNA. For the latter he received his second Np in chemistry in 1980, which was shared with Paul Berg (1926-, Stanford U.) and Walter Gilbert (1932-, Harvard U.) for their contributions, respectively, in recombinant DNA and chemical sequencing of DNA. The studies of insulin involved usage of DNP for tagging disulfide bonds as cysteic acid residues (1949), and paper chromatography introduced by Martin and Synge 1944. That it was the first elucidation of any protein structure lowered the barrier for future structure studies of proteins.

Stanford Moore, 1913-1982, Ph.D. Wisconsin (K.P. Link), Np 1972; Rockefeller, and William Stein, 1911-1980, Ph. D. Columbia (E.G. Miller); Rockefeller, Np 1972. Moore and Stein cooperatively developed methods for the rapid quantification of protein hydrolysates by combining partition chromatography, ninhydrin coloration and drop-counting fraction collector, i.e., the basis for commercial amino acid analyzers, and applied them to analysis of the ribonuclease structure.

Bruce Merrified, 1921-, Ph.D. UCLA (M. Dunn); Rockefeller, Np 1984. The concept of solid phase peptide synthesis using porous beads, chromatographic columns, and sequential elongation of peptides and other chains revolutionized the synthesis of biopolymers.

High performance liquid chromatography (HPLC), introduced around the mid-1960s and now coupled on-line to many analytical instruments, e.g., UV, FTIR, MS, is an indispensable daily tool found in all natural products chemistry laboratories.

Structures of Organic Compounds, 19th Century.

The discoveries made from 1848 to 1874 by Pasteur, Kekulé, van't Hoff, Le Bel and others led to a revolution in structural organic chemistry. Louis Pasteur (1822-95) was puzzled about why the potassium salt of tartaric acid (deposited on wine casks during fermentation) was dextrorotatory while the sodium ammonium salt of racemic acid (also deposited on wine casks) was optically inactive although both tartaric acid and "racemic" acid had identical chemical compositions. In 1848, the 25 year old Pasteur examined the racemic acid salt under the microscope and found two kinds of crystals exhibiting a left and right hand relation. Upon separation of the left-handed and right-handed crystals, he found that they rotated the plane of polarized light in opposite directions. He had thus performed his famous resolution of a racemic mixture, and had demonstrated the phenomenon of chirality. Pasteur went on to show that the racemic acid formed two kinds of salts with optically active bases such as quinine, i.e., this was the first demonstration of diastereomeric resolution. From this work Pasteur concluded that tartaric acid must have an element of asymmetry within the molecule itself. However, a three-dimensional understanding of the enantiomeric pair was only solved 25 years later (below). Pasteur's own interest shifted to microbiology where he made the crucial discovery of the involvement of "germs" or microorganisms in various processes and proved that yeast induces alcoholic fermentation, while other microorganisms lead to diseases; he thus saved the wine industries of France, originated the process known as *pasteurization*, and later developed vaccines for rabies. He was a genius who made many fundamental discoveries in chemistry and in microbiology.

The structures of organic compounds were still totally mysterious. Although Wöhler had synthesized urea, an isomer of ammonium cyanate, in 1828, the structural difference between those isomers was not known. In 1858 August Kekulé (1829-96; studied with André Dumas and C.A. Wurtz in Paris, taught at Ghent, Heidelberg and Bonn) published his famous paper in Liebig's Annalen der Chemie on the structure of carbon, in which he proposed that carbon atoms could form C-C bonds with hydrogen and other atoms linked to them; his dream on the top deck of a London bus led him to this concept. It was Butlerov who introduced the term "structure theory" in 1861. Further, in 1865 Kekulé conceived the cyclo-hexa-1:3:5-triene structure for benzene (C_6H_6) from a dream of a snake biting its own tale. In 1874, two young chemists, van't Hoff (1852-1911, Np 1901) in Utrecht, and Le Bel (1847-1930) in Paris, who had met in 1874 as students of C.-A. Wurtz, Paris, published the revolutionary 3D structure of the tetrahedral carbon Cabcd to explain the enantiomeric behavior of Pasteur's salts. The model was welcomed by J. Wislicenus (1835-1902; Zürich, Würzburg, Leipzig) who in 1863 had demonstrated the enantiomeric nature of the two lactic acids found by Scheele in sour milk (1780) and by Berzelius in muscle tissue (1807). This model, however, was criticized by Hermann Kolbe (1818-1884, Leipzig) as an "ingenious but in reality trivial and senseless natural philosophy." After 10 years of heated controversy, the idea of tetrahedral carbon was fully accepted, Kolbe had died and Wislicenus succeeded him in Leipzig.

Emil Fischer (1852-1919, Np 1902) was the next to make a critical contribution to stereochemistry. From the work of van't Hoff and LeBel he reasoned that glucose should have 16 stereoisomers. Fischer's doctorate work on hydrazines under Baeyer (1835-1917, Np 1905) at Strasbourg had led to studies of osazones which culminated in the brilliant establishment, including configurations, of the Fischer sugar tree starting from D-(+)-glyceraldehyde all the way up to the aldohexoses, allose, altrose, glucose, mannose, gulose, idose, galactose and talose (from 1884 to 1890). Unfortunately Fischer suffered from the toxic effects of phenylhydrazine for 12 years. The arbitrarily but luckily chosen absolute configuration of D-(+)-glyceraldehyde was luckily shown to be correct sixty years later in 1951 (Johannes-Martin Bijvoet, 1892-1980). Fischer's brilliant correlation of the sugars comprising the Fischer sugar tree was performed using the Kiliani (1855-1945)-Fischer method via cyanohydrin intermediates for elongating sugars. Fischer also made remarkable contributions to the chemistry of amino acids and to nucleic acid bases (see below).

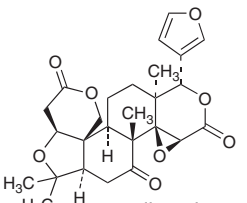
Structures of Organic Compounds, 20th Century.

The early concept of covalent bond was provided with a sound theoretical basis by Linus Pauling (1901-94, Np 1954), one of the greatest intellects of this century. Pauling's totally interdisciplinary research interests, including proteins and DNA is responsible for our present understanding of molecular structures. His books

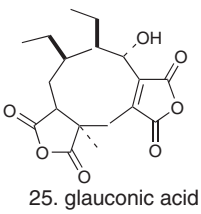
"Introduction to Quantum Mechanics" (with graduate student E.B. Wilson, 1935) and "The Nature of the Chemical Bond" (1939) have had a profound effect on our understanding of all of chemistry.

The actual 3D shapes of organic molecules which were still unclear in the late 1940s were then brilliantly clarified by Odd Hassel (1897-1981, Oslo U., Np 1969) and Derek Barton, (1918-1998, Np 1969). Hassel, an X-ray crystallographer and physical chemist, demonstrated by electron diffraction that cyclohexane adopted the chair form in the gas phase and that it had two kinds of bonds, "standing (axial)" and "reclining (equatorial)" (1943). Because of the German occupation of Norway in 1940, instead of publishing the result in German journals, he published it in a Norwegian journal which was not abstracted in English until 1945. During his 1949 stay at Harvard, Barton attended a seminar by Louis Fieser on steric effects in steroids and showed Fieser that interpretations could be simplified if the shapes (i.e., "conformations") of cyclohexane rings were taken into consideration; Barton made these comments because he was familiar with Hassel's study on cis- and trans-decalins. Following Fieser's suggestion Barton published these ideas in a 4 page *Experientia* paper (1950). This led to the joint Nobel prize with Hassel (1969), and established the concept of conformational analysis, which has exerted a profound effect in every field involving organic molecules.

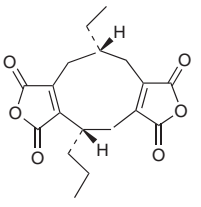
Using conformational analysis, Barton determined the structures of many key terpenoids such as β-amyrin, lanosterol, cycloartenone and cycloartenol (Birkbeck College). At Glasgow (from 1955) he collaborated in a number of cases with Monteath Robertson (1900-89) and established many challenging structures: limonin **24**, glauconic acid **25** and byssochlamic acid **26** ("nonadrides"). Barton was also associated with the Research Institute for Medicine and Chemistry (RIMAC), Cambridge, U.S.A. founded by the Schering company, where with J.M. Beaton, he produced 60 g of aldosterone at a time when the world supply of this important hormone was in mg quantities. Aldosterone **27** synthesis ("a good problem") was achieved in 1961 by Beaton ("a good experimentalist") through a nitrite photolysis, which came to be known as the Barton reaction ("a good idea")(quotes from his 1991 autobiography published by the American Chemical Society). From Glasgow, Barton went on to Imperial College, and a year before retirement, in 1977 he moved to France to direct the research at ICSN at Gif-sur-Yvette where he explored oxidation reactions selective for unactivated C-H. After retiring from ICSN he made a further move to Texas A & M in 1986, and continued his energetic activities, including chairman of the Tetrahedron publications. He felt weak during work one evening and died soon after, on March 16, 1998. He was fond of the phrase "gap jumping" by which he meant seeking generalizations between facts that do not seem to be related: "In the conformational analysis story, one had to jump the gap between steroids and chemical physics." (from his 1991 autobiography published by the American Chemical Society). According to Barton, the three most important qualities for a scientist are "intelligence, motivation, and honesty." His routine at Texas A & M was to wake around 4 am, read the literature, go to the office at 7 am and stay there until 7 pm; when asked in 1997 whether this was still the routine, his response was that he wanted to wake up earlier because sleep was a waste of time - a remark which characterized this active scientist approaching 80!



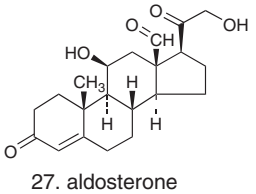
24. limonin
bitter principle of citrus fruits



25. glauconic acid
Penicillium glaucum
fatal livestock, poultry toxin



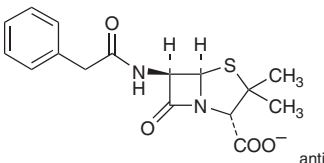
26. byssochlamic acid
Byssochlamys fulva
fatal livestock, poultry toxin



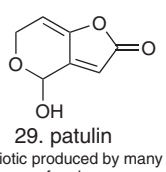
27. aldosterone
adrenocortical steroid
regulates metabolism of
minerals, water

Robert B. Woodward (1917-79, Np 1965), who died prematurely, is regarded by many as the preeminent organic chemist of this century. He made landmark achievements in spectroscopy, synthesis, structure determination, biogenesis, as well as in theory. His solo papers published in 1941-42 on empirical rules for estimating the absorption maxima of enones and dienes made the general organic chemical community realize that UV could be used for structural studies, thus launching the beginning of the spectroscopic revolution which soon brought on the applications of IR, NMR, MS, etc. He determined the structures of the following compounds: penicillin **28** in 1945 (through joint UK-USA collaboration, see Hodgkin), strychnine **2** in '48,

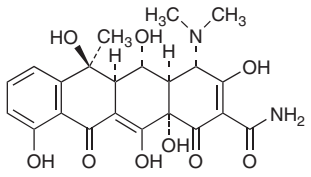
patulin **29** in '49, terramycin **30**, aureomycin **31** and ferrocene **32** (with G. Wilkinson, Np 1973 - shared with E. O. Fischer for sandwich compounds) in '52, cevine **33** in '54 (with Barton Np 1966, Jeger and Prelog, Np 1975), carbomycin **34** in '56, gliotoxin **35** in '58, oleandomycin **36** in '60, streptonigrin **37** in '63, and tetrodotoxin **10** in '64.



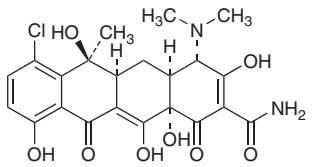
28. penicillin G salt
antibacterial



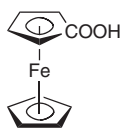
29. patulin
antibiotic produced by many fungi



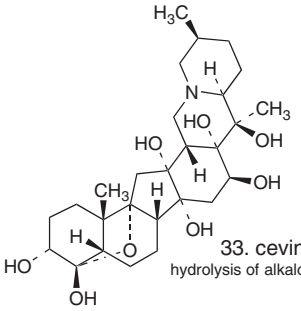
30. terramycin
Streptomyces rimosus
antibacterial



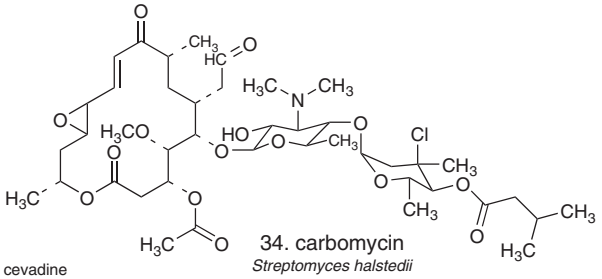
31. aureomycin
Streptomyces aureofaciens
antibacterial



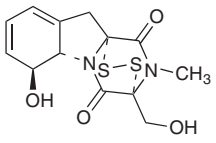
32. ferrocene
gasoline antiknock additive



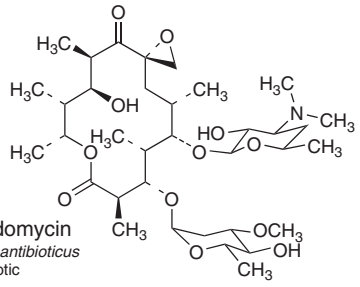
33. cevine
hydrolysis of alkaloid cevadine



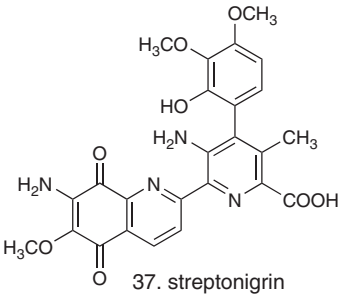
34. carbomycin
Streptomyces halstedii
asntibacterial



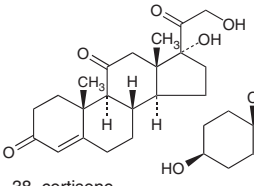
35. gliotoxin
antibiotic produced by *Pen.*, *Asper.*, etc.



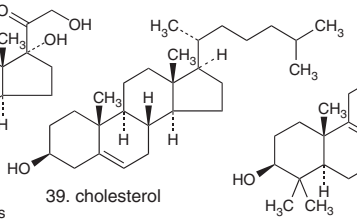
36. oleandomycin
Streptomyces antibioticus
antibiotic



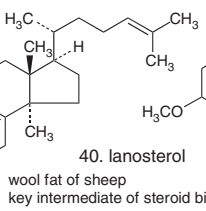
37. streptonigrin
Streptofloccului
antineoplastic



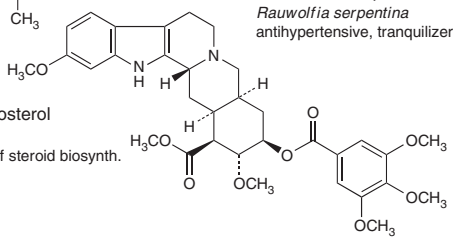
38. cortisone
from suprarenal glands
antiinflammatory



39. cholesterol

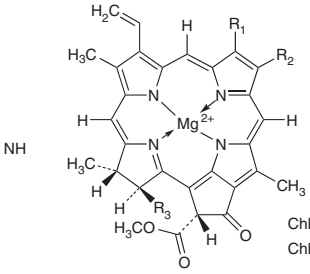


40. lanosterol
wool fat of sheep
key intermediate of steroid biosynth.



41. reserpine
Rauwolfia serpentina
antihypertensive, tranquilizer

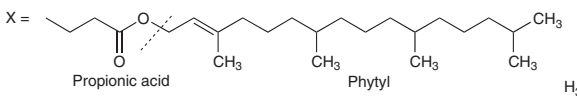
He synthesized patulin **29** in '50, cortisone **38** and cholesterol **39** in '51, lanosterol **40**, lysergic acid (see 21, with Eli Lilly) and strychnine **2** in '54, reserpine **41** in '56, chlorophyll **42** in '60, a tetracycline (with Pfizer) in '62, cephalosporin **43** in '65, and vitamin B_{12} **44** in '72 (with A. Eschenmoser, 1925- , ETH Zürich). He derived biogenetic schemes for steroids in '53 (with K. Bloch, see below), and for macrolides in '56, while the Woodward-Hoffmann orbital symmetry rules in '65 brought order to a large class seemingly random cyclization reactions.



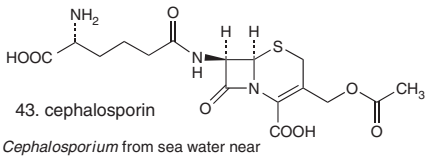
42. chlorophyll

green pigment of plants, algae deodorant

	R	R_2	R_3
Chlorophyll a	CH_3	CH_2CH_3	X
Chlorophyll b	CHO	CH_2CH_3	X

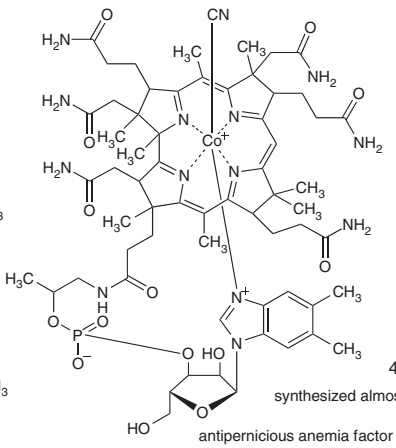


Propionic acid Phytyl



43. cephalosporin

Cephalosporium from sea water near sewage outlet on coast of Sardiani

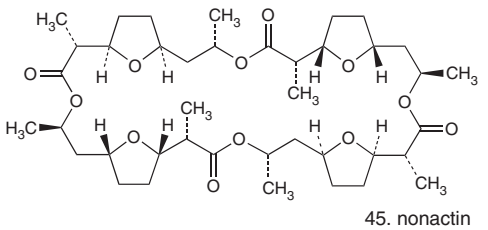


44. vitamin B_{12}

synthesized almost exclusively by bacteria

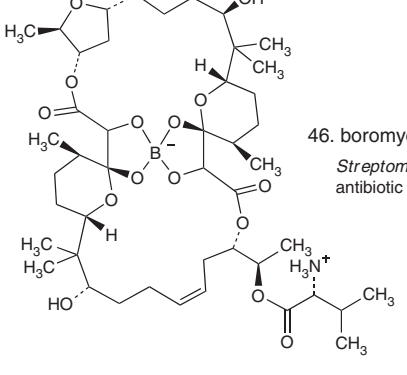
antipernicious anemia factor vitamin (hematopoietic)

Another central figure in stereochemistry is Vladimir Prelog (1906-1998, Np 1975), who succeeded Leopold Ruzicka at the ETH Zürich, and continued to build this institution into one of the most active and lively research and discussion centers in the world. The core group of intellectual leaders consisted of P. Plattner (1904-75), O. Jeger, A. Eschenmoser, J. Dunitz, D. Arigoni, and A. Dreiding (from Univ. Zürich). After completing extensive research on alkaloids, Prelog determined the structures of nonactin **45**, boromycin **46**, ferrioxamins **47** and rifamycins **48**. His seminal studies in the synthesis and properties of 8-12 membered ring led him into unexplored areas of stereochemisty and chirality. Together with Robert Cahn (1899-1981, London Chem. Soc.) and Christopher Ingold (1893-1970, U. College, London; pioneering mechanistic interpretation of organic reactions), he developed the Cahn-Ingold-Prelog (CIP) sequence rules for the unambiguous specification of stereoisomers. Prelog was an excellent story teller, always had jokes to tell, and was respected and loved by all who knew him.



45. nonactin

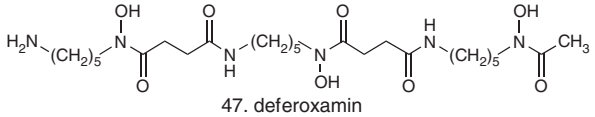
Streptomyces spp. antibiotic



46. boromycin

Streptomyces antibioticus antibiotic

antibiotic

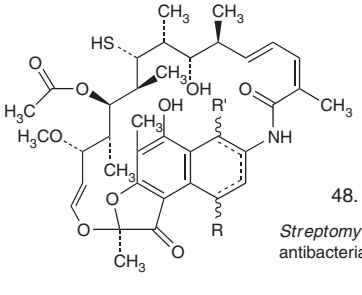


47. deferoxamin

Streptomyces pilosus

forms iron complexes

parenteral chelating agent



48. rifamycins

Streptomyces mediterranei

antibacterial (tuberculostatic)

	R	R'
Rifamycin B	$-OCH_2COOH$	—OH
Rifamycin O	—(1,3-dioxolan-4-on)-2-yl	=O
Rifamycin S	=O	=O
Rifamycin X	$=N^+=N^-$	=O

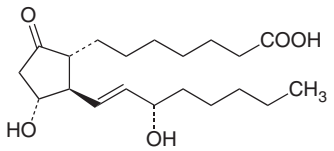
Polyketides, Fatty acids.

Arthur Birch (1915-95) from Sydney U., Ph. D. with Robert Robinson (Oxford U.), then professor at Manchester U. and Australian National U., was one of the earliest chemists to perform biosynthetic studies using radiolabels; starting with polyketides he studied the biosynthesis of a variety of natural products such as the C_6-C_3-C_6 backbone of plant phenolics, polyene macrolides, terpenoids, and alkaloids. He is especially known for the Birch reduction of aromatic rings, metal-ammonia reductions leading to 19-norsteroid hormones and other important products (1942-), which were of industrial importance. Feodor Lynen (1911-1979, Np 1964) performed studies on the intermediary metabolism of the living cell that led him to the demonstration of the first step in a chain of reactions resulting in the biosynthesis of sterols and fatty acids.

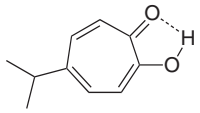
Prostaglandins **49**, a family of 20 carbon, lipid-derived acids discovered in seminal fluids and accessory genital glands of man and sheep by von Euler (1934), have attracted great interest because of their extremely diverse biological activities. They were isolated and their structures elucidated from 1963- on by S. Bergström (1916- , Np 1982) and B. Samuelsson (1934- , Np 1982) at the Karolinska Institute, Stockholm. Many syntheses of the natural prostaglandins and their non-natural analogs have been published.

Tetsuo Nozoe (1902-1996) who studied at Tohoku U., Sendai, with Riko Majima (1874-1962, see below) went to Taiwan where he stayed until 1948 before returning to Tohoku U. At National Taiwan U. he isolated hinokitiol **50** from the essential oil of taiwanhinoki. Remembering the resonance concept put forward by Pauling just before W.W.II, he arrived at the 7-membered nonbenzenoid aromatic structure for hinokitiol in 1941, the first of the troponoids. This highly original work remained unknown to the rest of the world until 1951. In the meantime, during 1945-1948, nonbenzenoid aromatic structures had been assigned to stipitatic acid **51** (isolated by H. Raistrick) by Michael J. S. Dewar (1918-1997) and to the thujaplicins **52** by Holger Erdtman (1902-1989); the term tropolones was coined by Dewar in 1945. Nozoe continued to work on and discuss troponoids, up to the night before his death, without knowing that he had cancer. He was a remarkably focused and warm scientist, working all the time. Erdtman (Royal Inst. Techn. Stockholm) was the central figure in Swedish natural products chemistry who, with his wife Gunhild Aulin Erdtman (dynamic, General Secretary of the Swedish Chem. Soc.), worked in the area of plant phenolics.

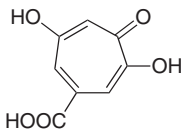
As mentioned in the following and in the concluding sections, classical biosynthetic studies using radioactive isotopes for determining the distribution of isotopes has now largely been replaced by the use of various stable isotopes coupled with NMR and MS. The main effort has now shifted to the identification and cloning of genes, if possible the gene clusters, involved in the biosynthesis of the natural product. In the case of polyketides (acyclic, cyclic and aromatic), the focus is on the polyketide synthases.



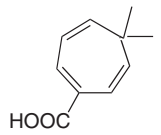
49. prostglandin E_1
sheep seminal vesicle tissue
vasodilator



50. hinokitiol
hinoki (a cedar)



51. stipitatic acid
Penicillium. stipitatum



52. thujic acid
Thuja plicata (cedar)
antibiotic

First nonbenzenoid aromatics

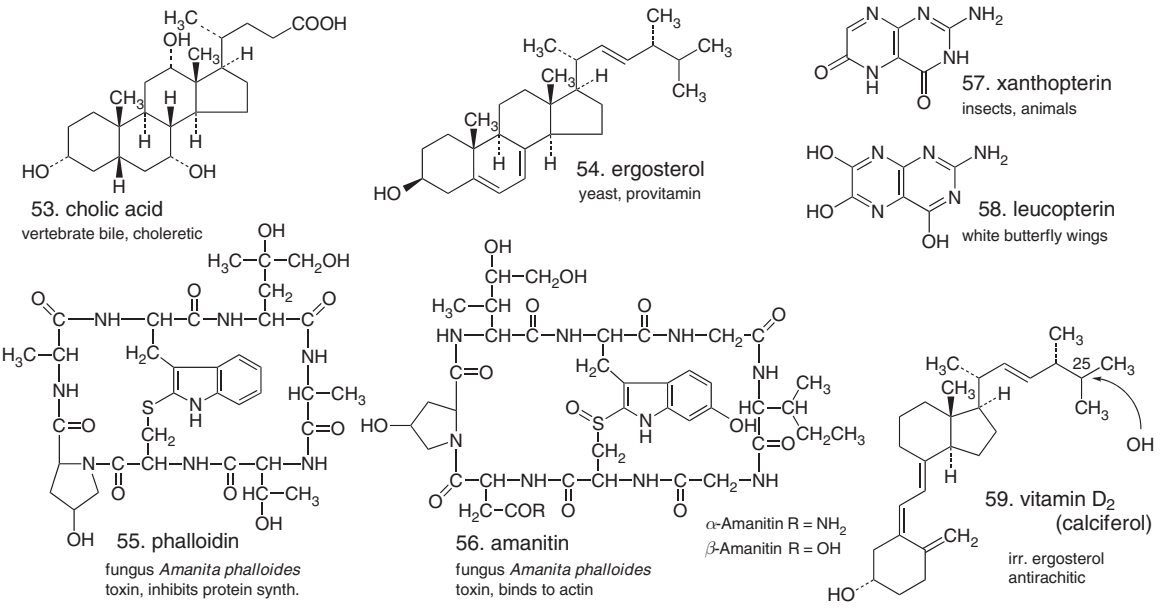
Isoprenoids, steroids, carotenoids.

During his assistantship to Kekulé at Bonn, Otto Wallach (1847-1931, Np 1910) had to familiarize himself with the essential oils from plants; many of the components of these oils were compounds for which no structure was known. In 1891 he clarified the relations between 12 different monoterpenes related to pinene. This was summarized together with other terpene chemistry in book form in 1909, and led him to propose the "isoprene rule." These achievements laid the foundation for the future development of terpenoid chemistry and brought order from chaos.

The next period up to around 1950 saw phenomenal advances in natural products chemistry centered on isoprenoids. Many of the best natural products chemists in Europe, including Wieland, Windaus, Karrer, Kuhn, Butenandt, Ruzicka contributed to this breathtaking pace. Heinrich Wieland (1877-1957) worked on the bile acid

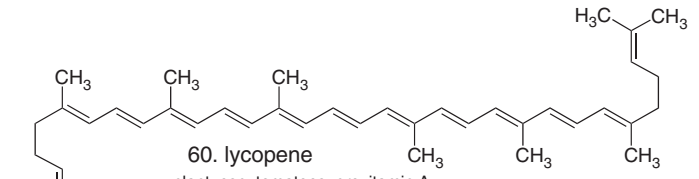
53 structure, which had been studied over a period of 100 years and considered to be one of the most difficult to attack; he received the Nobel Prize in 1927 for these studies. On the other hand, his friend Adolph Windaus (1876-1959) worked on the structure of cholesterol for which he also received the Nobel Prize in 1928.

Unfortunately, there were chemical discrepancies in the proposed steroidal skeletal structure, which had a 5-membered ring B attached to C-7 and C-9. J.D. Bernal, Mineralogical Museums, Cambridge U., who was examining the X-ray patterns of ergosterol **54** (1932) noted that the dimensions were inconsistent with the Wieland-Windaus formula. A reinterpretation of the production of chrysene from sterols by Pd/C dehydrogenation reported by Diels (see below) in 1927 eventually led Rosenheim/King and Wieland/Dane to deduce the correct structure in 1932. Wieland also worked on the structures of morphine / strychnine alkaloids, phalloidin **55**/ amanitin **56** cyclopeptides of toxic mushroom *Amanita phalloides*, and pteridines **57**/**58**, the important fluorescent pigments of butterfly wings. Windaus determined the structure of ergosterol and continued structural studies of its irradiation product which exhibited antirachitic activity "vitamin D" **59**. The mechanistically complex photochemistry of ergosterol leading to the vitamin D group has been investigated in detail by Egbert Havinga (1927-1988, Leiden U.), a leading photochemist and excellent tennis player.

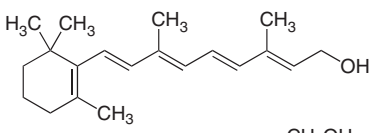


Paul Karrer (1889-1971, Np 1937), established the foundations of carotenoid chemistry through structure determinations of lycopene **60** and other carotenoids, vitamin A **61**, etc., and the synthesis of squalene, carotenoids and others. George Wald (1906-97, Np 1967) showed that vitamin A was the key compound in vision while stayring in Karrer's laboratory. Vitamin K **62** (K from "Koagulation"), discovered by Henrik Dam (1895-1976, Polytechnic Inst., Copenhagen, Np 1943) and structurally studied by Edward Doisy (1893-1986, St. Louis U., Np 1943), was also synthesized by Karrer. In addition, Karrer synthesized riboflavin **63** (vitamin B_2) and determined the structure and role of nicotinamide adenine dinucleotide phosphate **64** ($NADP^+$) with Otto Warburg.

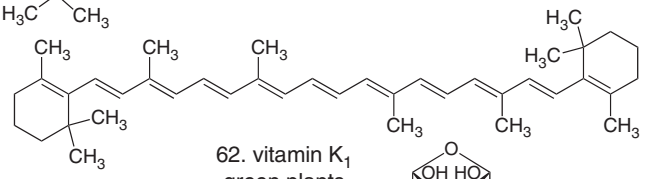
The research on carotenoids and vitamins of Karrer who was at the U. of Zürich overlapped with that of Richard Kuhn (1900-1967, Np 1938) at the ETH Zürich, and the two were frequently rivals. Richard Kuhn, one of the pioneers in using UV/vis spectroscopy for structural studies, i.e., carotenoids, introduced the concept of "atropisomerism" in diphenyls, and studied the spectra of series of diphenyl polyenes. He determined structures of many natural carotenoids, proved the structure of riboflavin-5-phosphate (flavin-adenine-dinucleotide-5-phosphate **65**) and showed that the combination of NAD-5-phosphate with the carrier protein yielded the yellow oxidation enzyme, thus providing an understanding of the role of a prosthetic group. He also determined the structures of vitamin B complex, i.e., pyridoxine **66**, p-aminobenzoic acid **67**, pantothenic acid **68** after W.W.II he went on to structural studies of N-containing oligosaccharides in human milk that provides immunity for infants, and brain gangliosides. Carotenoid studies in Switzerland were later taken up by Otto Isler (1910-93), a Ruzicka student, at Hoffmann-La Roche and Conrad Hans Eugster (1921-), a Karrer student at Zurich U.



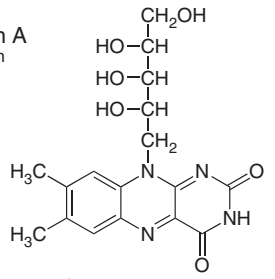
60. lycopene
plant, esp. tomatoes; provitamin A



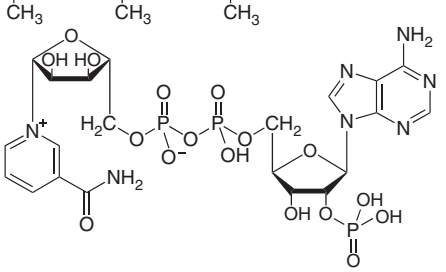
61. vitamin A
animals, fish



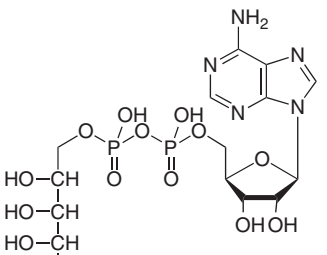
62. vitamin K_1
green plants

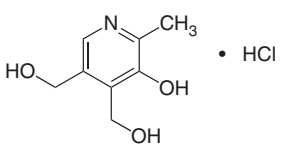


63. riboflavin, vit. B_2
milk, egg, kidney, leafy vegetables
nutritional factor

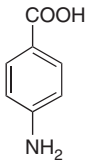


64. nicotinamide adenine dinucleotide phosphate, NADP
coenzyme of dehydrogenases

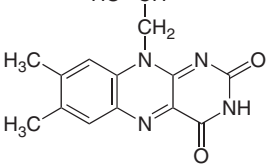




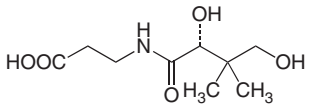
66. pyridoxine hydrochloride
yeast, liver, foodstuff
member of Vit. B6 complex;



67. p-aminobenzoic acid
yeast, widely in nature
bacterial vit., anticanitic vit.; UV screen



65. flavine-adenine-dinucleotide (FAD)
prosthetic group of flavoenzymes
yeast, liver, kidney heart, muscle

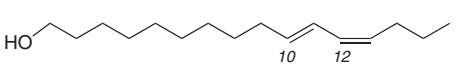


68. pantothenic acid
all animal & plant tissues
essential vit. for CO-A biosynth.

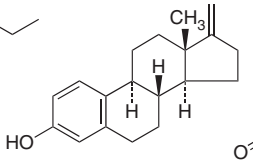
Adolf Butenandt (1903-1998, Np 1939) initiated and essentially completed isolation and structural studies of the human sex hormones, the insect molting hormone (ecdysone) and the first pheromone, bombykol **69**. With help from industry he was able to obtain large supplies of urine of pregnant women for estrone **70**, sow ovaries for progesterone **71**, and 4,000 gallons of male urine for androsterone **72** (50 mg, crystals), he isolated and determined structures of two female sex hormones, estrone and progesterone, and the male hormone androsterone all during the period 1934-39 (!) and was awarded the Nobel prize in 1939. Keen intuition, use of UV data and Pregl's microanalysis all played important roles. He was appointed to a professorship in Danzig at age 30. With Peter Karlson he isolated from 500 kg of silkworm larvae 25 mg of α-ecdysone **73**, the prohormone of insect and crustacean molting hormone, and determined its structure as a polyhydroxysteroid (1965); 20-hydroxylation gives the insect and crustacean molting hormone or β-ecdysone **74** (20-hydroxyecdysteroid). He also was the first to isolate an insect pheromone, bombykol from female silkworm moths (with E. Hecker). As president of the Max Planck Foundation, he strongly influenced the postwar rebuilding of German science.

The successor to Kuhn, who left ETH Zürich for Heidelberg, was Leopold Ruzicka (1887-1967, Np 1939) who established a close relation with the Swiss pharmaceutical industry. His synthesis of the 17- and 15-membered macrocyclic ketones, civetone **75** and muscone **76** (the constituents of musk) showed that contrary to Baeyer's prediction, large alicyclic rings could be strainless. He reintroduced and refined the isoprene rule

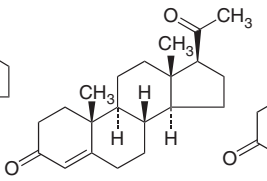
proposed by Wallach (1887) and determined the basic structures of many sesqui-, di- and triterpenes, as well as the structure of lanosterol, the key intermediate in cholesterol biosynthesis. The "biogenetic isoprene rule" of the ETH group, Albert Eschenmoser, Leopold Ruzicka, Oskar Jeger and Duilio Arigoni contributed to a concept of terpenoid cyclization (1955), which was consistent with the mechanistic considerations put forward by Stork as early as 1950. Besides making the ETH group into a center of natural products chemistry, Ruzicka bought many 17th century Dutch paintings with royalties accumulated during the war from his Swiss and American patents, and donated them to the Zürich Kunsthaus.



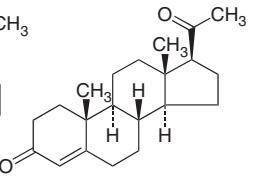
69. bombykol
Bombyx mori (silk-worm)
first insect pheromone (1959)



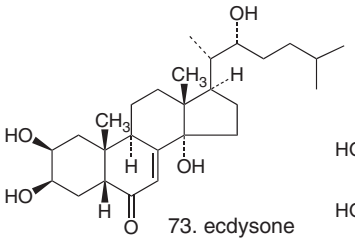
70. estrone
pregnancy urine of
women estrogen



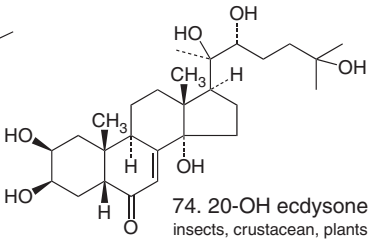
71. progesterone
progestogen
corpus luteum



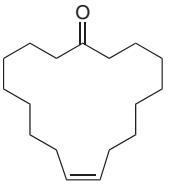
72. androsterone
male urine



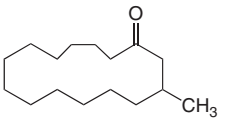
73. ecdysone
insects, crustacean, plants
molting prohormone



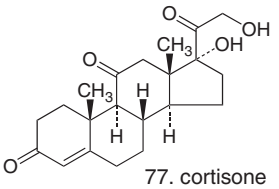
74. 20-OH ecdysone
insects, crustacean, plants
molting hormone



75.civetone
civet
perfumery



76.muscone
musk
perfumery

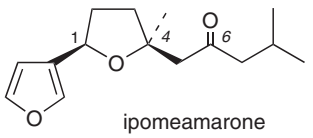


77. cortisone
suprarenal gland
antiinflammatory

Studies in the isolation, structures, and activities of the antiarthritic hormone, cortisone **77** nd related compounds from the adrenal cortex were performed in the mid- to late 1940s during W.W.II. by Edward Kendall (1886-1972, Mayo Clinic, Rochester, Np 1950), Tadeus Reichstein (1897-1996, Basel U., Np 1950), Philip Hench (1896-1965, Mayo Clinic, Rochester, Np 1950), Oskar Wintersteiner (1898-1971, Columbia U., Squibb) and others initiated interest as an adjunct to military medicine as well as to supplement the meager supply from beef adrenal glands by synthesis. Lewis Sarett (1917-, Merck & Co., later president) and coworkers completed the cortisone synthesis in 28 steps, one of the first two totally stereocontrolled synthesis of a natural product; the other was cantharidin, Stork 1951 (see above). The multistep cortisone synthesis was put on the production line by Max Tishler (1906-89, Merck & Co , later president) who made contributions to the synthesis of a number of drugs, including riboflavin. Besides working on steroid reactions/synthesis and antimalarial agents, Louis F. Fieser (1899-1977) and Mary Fieser (1909-1997) of Harvard U. made huge contributions to the chemical community through their outstanding books: *Natural Products related to Phenanthrene* (1949), *Steroids* (1959), *Advanced Organic Chemistry* (1961), *Topics in Organic Chemistry* (1963) as well as their textbooks and an important series of books on Organic Reagents. Carl Djerassi (1923-, Stanford U.), a prolific chemist, industrialist, and more recently a novelist, started to work at the Syntex laboratories in Mexico City where he directed the work leading to the first oral contraceptive ("the pill") for women; Djerassi also re-introduced optical rotatory dispersion (ORD, see below) and was also a pioneer in organic mass spectgrometry.

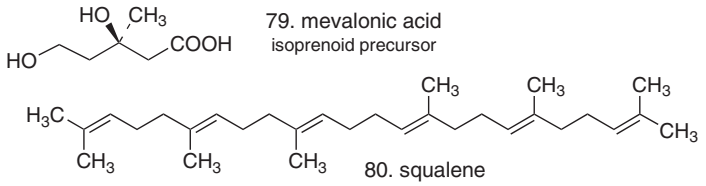
Takashi Kubota (1909- , Osaka City U.), with Teruo Matsuura (1924-, Kyoto U.), determined the structure of the furanoid sesquiterpene, ipomeamarone **78**, from the black rotted portion of spoiled sweet potatoes; this

research constitutes the first characterization of a phytoallexin, defense substances produced by plants in response to attack by fungi or physical damage. Damaging a plant and characterizing the defense substances produced may lead to new bioactive compounds. The mechanism of induced biosynthesis of phytoallexins, which is not fully understood, is an interesting biological mechanistic topic that deserves further investigation. Another center of high activity in terpenoids and nucleic acids was headed by Frantisek Sorm (1913-80), Inst. Organic and Biochemistry, Prague, who determined the structures of many sesquiterpenoids and other natural products; he was not only active scientifically but also was a central figure who helped to guide the careers of many Czech chemists.



78. ipomeamarone
black rotten sweet potato
first phytoallexin

The key compound in terpenoid biosynthesis is mevalonic acid **79** (MVA) derived from acetyl-CoA, which was discovered fortuitously in 1957 by the Merck team in Rahway N.J. headed by Karl Folkers (1906-98). They soon realized and proved that this C_6 acid was the precursor of the C_5 isoprenoid unit isopentenyl diphosphate (IPP) that ultimately leads to the biosynthesis of cholesterol. In 1952 Konrad Bloch (1912-, Harvard, Np 1964) with R.B. Woodward published a paper suggesting a mechanism of the cyclization of squalene **80** to lanosterol and the subsequent steps to cholesterol, which turned out to be essentially correct. This biosynthetic path from MVA to cholesterol was experimentally clarified in stereochemical detail by John Cornforth (1917-, Np 1975) and George Popják. In 1932, Harold Urey (1893-1981, Np 1934) of Columbia U. discovered heavy hydrogen. Urey showed, contrary to common expectation, that isotope separation could be achieved with deuterium in the form of deuterium oxide by fractional electrolysis of water. Urey's separation of the stable isotope deuterium led to the isotopic tracer methodology that revolutionized the protocols for elucidating biosynthetic processes and reaction mechanisms, as exemplified beautifully by the cholesterol studies. Using MVA labeled chirally with isotopes, including chiral methyl, i.e., -CHDT, Cornforth and Popják clarified the key steps in the intricate biosynthetic conversion of mevalonate to cholesterol in stereochemical detail. The chiral methyl group was also prepared independently by Duilio Arigoni (1928-, ETH, Zürich). Cornforth has had great difficulty in hearing and speech since childhood but has been helped expertly by his chemist wife Rita; he is an excellent tennis and chess player, and is renowned for his speed in composing witty occasional limericks.

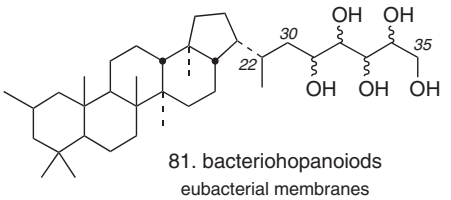


79. mevalonic acid
isoprenoid precursor

80. squalene
shark liver oil

Although MVA has long been assumed to be the only natural precursor for IPP, a non-MVA pathway in which IPP is formed via the glyceradehyde phosphate-pyruvate pathway has been discovered (1995-96) in the ancient bacteriohopanoids **81** by Michel Rohmer, who started working on them with Guy Ourisson (1926-,U. Strasbourg, terpenoid studies, including prebiotic), and by Duilio Arigoni in the ginkgolides, which are present in the ancient *Ginkgo biloba* tree. It is possible that many other terpenoids are biosynthesized via the non-MVA route. In classical biosynthetic experiments, ^{14}C-labeled acetic acid was incorporated into the microbial or plant product, and location

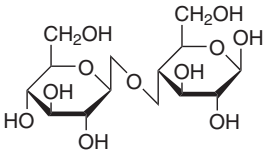
or distribution of the ^{14}C label was deduced by oxidation or degradation to specific fragments including acetic acid; therefore it was not possible or extremely difficult to map the distribution of all radioactive carbons. The progress in ^{13}C NMR made it possible to incorporate ^{13}C labeled acetic acid and locate all labeled carbons. This led to the discovery of the nonmevalonate pathway leading to the IPP units. Similarly, NMR and MS have made it possible to use the stable isotopes, e.g., ^{18}O, ^{2}H, ^{15}N, etc. in biosynthetic studies. The current trend of biosynthesis has now shifted to genomic approaches for cloning the genes of various enzyme synthases involved in the biosynthesis.



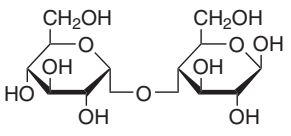
81. bacteriohopanoiods
eubacterial membranes
1–12 tons in fossil hopanoids

Carbohydrates, cellulose.

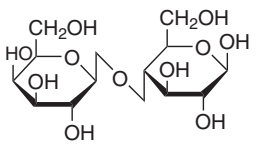
The most important advance in carbohydrate structures following those made by Emil Fischer was the change from acyclic to the current cyclic structure introduced by Walter Haworth (1883-1937). He noticed the presence of α- and β-anomers, and determined the structures of important disaccharides including cellobiose **82**, maltose **83**, and lactose **84**. He also determined the basic structural aspects of starch, cellulose, inulin and other polysaccharides, and accomplished the structure determination and synthesis of vitamin C **85**, a sample of which he had received from Albert von Szent-Györgyi (1893-1986, Np 1937). This first synthesis of a vitamin was significant since it showed that a vitamin could be synthesized in the same way as any other organic compound. There was strong belief among leading scientists in the 1910s that cellulose, starch, protein, and rubber were colloidal aggregates of small molecules. However, Hermann Staudinger (1881-1965, Np 1953) who succeeded R. Willstätter and H. Wieland at the ETH Zürich and Freiburg, respectively, showed through viscosity measurements and various molecular weight measurements that macromolecules do exist, and developed the principles of macromolecular chemistry.



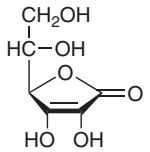
82. cellobiose
4-(β-D-glucosido)-D-glucose
unit of cellulose



83. maltose
4-(α-D-glucosido)-D-glucose
sweetener



84. lactose
4-(β-D-galactosido)-D-glucose
in mammalian milk



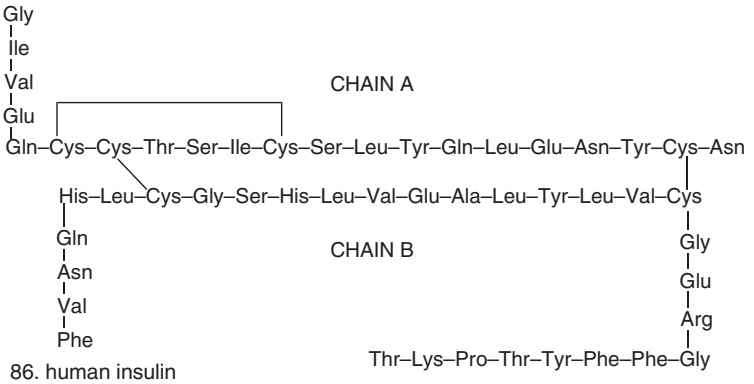
85. vitamin C
ascorbic acid
plants, animal

In more modern times, Raymond Lemieux (1920- , U. Ottawa and Alberta) has been a leader in carbohydrate research. He introduced the concept of endo- and exo-anomeric effects, accomplished the challenging synthesis of sucrose (1953), pioneered in the use of NMR coupling constants in configuration studies, and most importantly, starting with syntheses of oligosaccharides responsible for human blood group determinants, he prepared antibodies and clarified fundamental aspects of the binding of oligosaccharides by lectins and antibodies. The periodate-potassium permanganate cleavage of double bonds at room temperature (1955) is called the Lemieux reaction.

Amino acids, Peptides, Porphyrins and Alkaloids

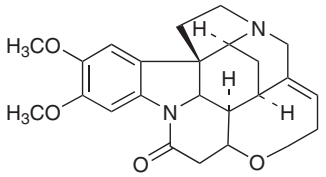
It is fortunate that we have China's record and practice of herbal medicine over the centuries, which is providing us with an indispensable source of knowledge. China is rapidly catching up in terms of infrastructure

and equipment in organic and bioorganic chemistry, and work on isolation, structure determination and synthesis stemming from these valuable sources has picked up momentum. However, as mentioned above, clarification of the active principles and mode of action of these plant extracts will be quite a challenge since in many cases synergistic action is expected. Wang Yu (1910-1997) who headed the well-equipped Shanghai Institute of Organic Chemistry surprised the world with the total synthesis of bovine insulin **86** performed by his group in 1965 ; the human insulin was synthesized around the same time by P. G. Katsoyannis, A. Tometsko and C. Zaut of the Brookhaven National Laboratory (1966).

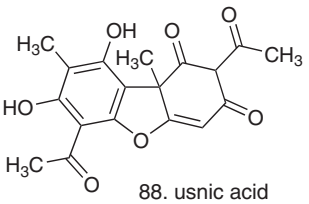


86. human insulin
produced by pancreatic beta cells
antidiabetic

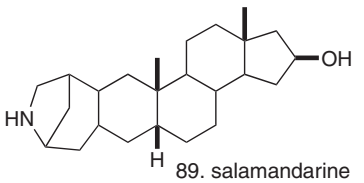
One of the giants in natural products chemistry during the first half of this century was Robert Robinson (1886-1975, Np 1947) at Oxford U. His synthesis of tropinone, a bicyclic amino ketone related to cocaine, from succindialdehyde, methylamine and acetone dicarboxylic acid under Mannich reaction conditions was the first biomimetic synthesis (1917). It reduced Willstätter's 1903 thirteen step synthesis starting with suberone into a single step. This achievement demonstrated Robinson's analytical prowess. He was able to dissect complex molecular structures into simple biosynthetic building blocks, and allowed him to propose the biogenesis of all types of alkaloids and other natural products. His laboratory at Oxford, where he developed the well-known Robinson annulation reaction (1937) in connection with his work on the synthesis of steroids became a world-center for natural products study. Robinson was a pioneer in the so-called electronic theory of organic reactions, and introduced the use of curly arrows to show the movements of electrons. His analytical power is exemplified in the structural studies of strychnine **2** and brucine **87** done in 1946-1952. Barton clarified the biosynthetic route to the morphine alkaloids, which he saw as an extension of his biomimetic synthesis of usnic acid **88** through a one-electron oxidation; this was later extended to a general phenolate coupling scheme. Morphine total synthesis was brilliantly achieved by Marshall Gates (1915-, U. Rochester) 1952.



87. brucine
Strychnos seeds
CNS stimulant



88. usnic acid
antibacterial in lichens

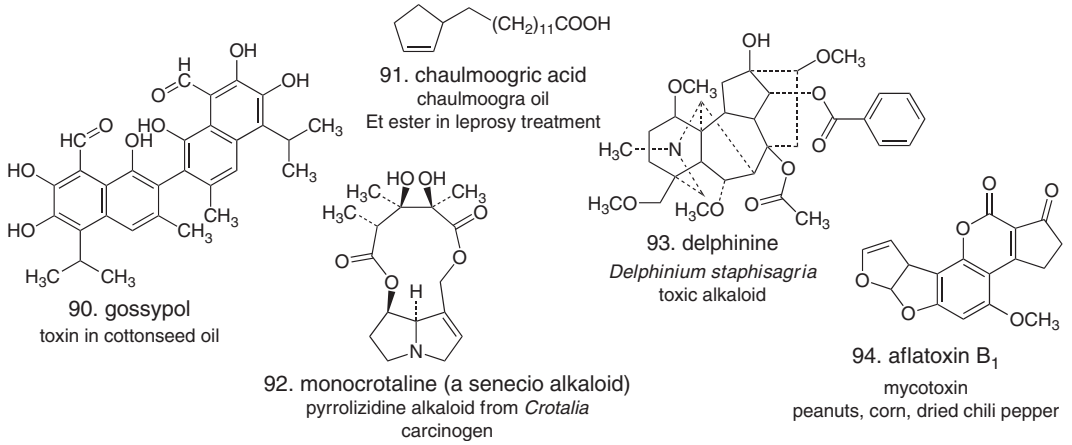


89. salamandarine
Salamander maculosa
toxin from glands near eye

The yield of the Robinson tropinone synthesis was low but Clemens Schöpf (1899-1970), Ph.D. Munich (Wieland), U. Darmstadt, improved it to 90% by carrying out the reaction in buffer; he also worked on the stereochemistry of morphine and determined the structure of the steroidal alkaloid salamandarine **89** (1961), the toxin secreted from glands behind the eyes of the salamander, a tailed amphibian.

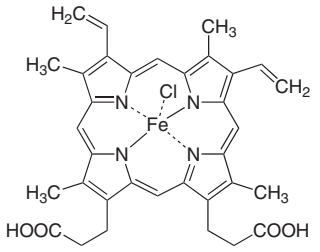
Roger Adams (1889-1971, U. Illinois), was the central figure in organic chemistry in the U.S.A. and is credited with contributing to the rapid development of its chemistry in the late 1930's and 40's, including training of graduate students for both academe and industry. After earning a Ph.D. in 1912 at Harvard U. he did postdoctoral studies with Otto Diels (see below) and Richard Willstätter (see below) in 1913; he once said that around those years in Germany he could cover all J. Am. Chem. Soc. papers published in a year in a single night. His important work include determination of the structures of tetrahydrocannabinol in marijuana, the toxic gossypol **90** in cottonseed oil, chaulmoogric acid **91** used in treatment of leprosy, and the *Senecio* alkaloids **92** with Nelson Leonard (1916-, U. Illinois, now at Caltech.). He also contributed to many fundamental organic reactions and syntheses. The famous Adams Pt catalyst are not only important for reducing double bonds in industry and in the laboratory, but was central for determining the number of double bonds in a structure. He was also one of the founders of the *Organic Synthesis* (started in 1921) and the *Organic Reactions* series. Nelson Leonard switched interests to bioorganic chemistry and biochemistry, where he has worked with nucleic acid bases and nucleotides, coenzymes, dimensional probes, and fluorescent modifications such as ethenoguanine.

The complicated structures of the medieval plant poisons aconitine **4** (from *Aconitum*) and delphinine **93**(from *Delphinium*) were finally characterized in 1959-60 by: Karel Wiesner (1919-1986. U. New Brunswick); Leo Marion (1899-1979, National Res. Council, Ottawa); George Büchi (1921-1998, mycotoxins, aflatoxin **94**/DNA adduct, synthesis of terpenoids and N-containing bioactive compounds, photochemistry); and Maria Przybylska (1923-, X-ray).

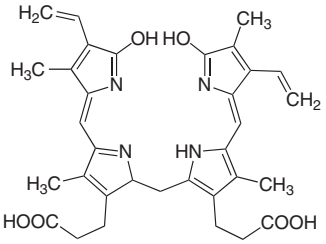


The complex chlorophyll structure was elucidated by Richard Willstätter (1872-1942, Np 1915). Although he could not join Baeyer's group at Münich because the latter had ceased taking students, a close relation developed between the two. During his chlorophyll studies, Willstätter re-introduced the important technique of column chromatography published in Russian by Michael Tswett (1906). Willstätter further demonstrated that Mg was an integral part of chlorophyll, clarified the relation between chlorophyll and the blood pigment hemin, and found the wide distribution of carotenoids in tomato, egg yolk and bovine corpus luteum. Willstätter also synthesized cyclooctatetraene and showed its properties to be wholly unlike benzene but close to those of acyclic polyenes (around 1913). He succeeded Baeyer at Münich in 1915, synthesized the anesthetic cocaine, retired early in protest of anti-Semitism, but remained active until the Hitler era, and in 1938 emigrated to Switzerland.

The hemin structure was determined by another German chemist of the same era, Hans Fischer (1881-1945, Np 1930), who succeeded Windaus at Innsbruck and at Münich. He worked on the structure of hemin **95** from the blood pigment hemoglobin, and completed its synthesis in 1929. He continued Willstätter's structural studies of chlorophyll, and further synthesized bilirubin **96** in 1944. Destruction of his institute at Techn. Hochsch. Münich, during W.W.II. led him to take his life in March, 1945. The biosynthesis of hemin was elucidated largely by David Shemin (1911-1991).



95. hemin
from hemoglobin



96. bilirubin
in bile and serum chrom.
involved in jaundice

In the mid 1930s the Department of Biochemistry at Columbia Medical School, which had accepted many refugees from the Third Reich, including Erwin Chargaff, Rudolf Schoenheimer and others on the faculty, and Konrad Bloch (see above) and David Shemin as graduate students, was a great center of research activity. In 1940, Shemin ingested 66 g of ^{15}N-labeled glycine over a period of 66 hours in order to determine the half-life of erythrocytes. David Rittenberg's analysis of the heme moiety with his home-made mass spectrometer showed all four pyrrole nitrogens came from glycine. Using ^{14}C (that had just become available) as a second isotope (see next paragraph), doubly labeled glycine $^{15}NH_2^{14}CH_2COOH$ and other precursors, Shemin showed that glycine and succinic acid condensed to yield δ-aminolevulinate, thus elegantly demonstrating the novel biosynthesis of the porphyrin ring (around 1950). At this time, Bloch was working on the other side of the bench.

Melvin Calvin (1911-1997, Np 1961) at UC Berkeley, elucidated the complex photosynthetic pathway in which plants reduce carbon dioxide to carbohydrates. The critical $^{14}CO_2$ had just been made available at UC Berkeley Lawrence Radiation Laboratory as a result of the pioneering research of Martin Kamen (1913-), while paper chromatography also played crucial roles. Kamen produced ^{14}C with Sam Ruben (1940), used ^{18}O to show that oxygen in photosynthesis comes from water and not from carbon dioxide, participated in the Manhattan project, testified before the House UnAmerican Activities Committee (1947), won compensatory damages from the US Department of State, and helped build the U. Calif. La Jolla (1957). The entire structure of the photosynthetic reaction center (>10,000 atoms) from the purple bacterium *Rhodopseudomonas viridis* has been established by X-ray crystallography in the landmark studies performed by Johann Deisenhofer, (1943-), Robert Huber (1937-) and Hartmut Michel (1948-) in 1989; this was the first membrane protein structure determined by X-ray, for which they shared the 1988 Nobel prize. The information gained from the full structure of this first membrane protein has been especially rewarding.

The recent studies on vitamin B_{12}, the structure of which was established by crystallographic studies performed by Dorothy Hodgkin (1910-94, Np 1964) is fascinating. Hodgkin also determined the structure of penicillin (in a joint effort between UK and American scientists during W.W.II) and insulin. The formidable total synthesis of vitamin B_{12} was completed in 1972 through collaborative efforts between Woodward and Eschenmoser, involving 100 postdoctoral fellows and extending over 10 years. The biosynthesis of fascinating complexity is almost completely solved through studies performed by Alan Battersby (1925-, Cambridge U.), Duilio Arigoni, and Ian Scott (1928-, Texas A & M) and collaborators where advanced NMR techniques and synthesis of labeled precursors is elegantly combined with cloning of enzymes controlling each biosynthetic step. This work provides a beautiful demonstration of the power of the combination of bioorganic chemistry, spectroscopy and molecular biology, a future direction which will become increasingly important for the creation of new "unnatural" natural products.

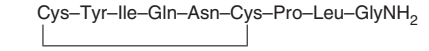
Enzymes, Proteins.

In the early days of natural products chemistry, enzymes and viruses were very poorly understood. Thus, the 1926 paper by James Sumner (1887-1955) at Cornell U. on crystalline urease was received with ignorance or skepticism, especially by Willstätter who believed that enzymes were small molecules and not proteins. John Northrop

(1891-1987) and co-workers at the Rockefeller Institute went on to crystallize pepsin, trypsin, chymotrypsin, ribonuclease, deoyribonuclease, carboxypeptidase and other enzymes between 1930-35. Despite this, for many years biochemists did not recognize the significance of these findings, and considered enzymes as being low molecular weight compounds adsorbed onto proteins or colloids. Using Northrop's method for crystalline enzyme preparations, Wendell Stanley (1904-71) at Princeton obtained tobacco mosaic virus as needles from one ton of tobacco leaves (1935). Sumner, Northrop and Stanley shared the 1946 Nobel prize in chemistry. All these studies opened a new era for biochemistry.

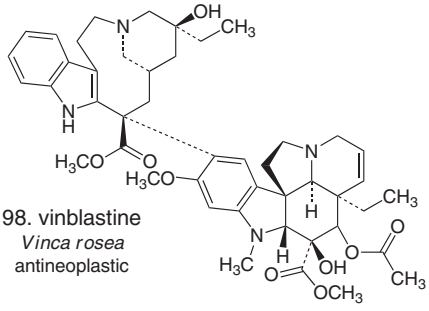
Meanwhile, Linus Pauling, who in mid-1930 became interested in the magnetic properties of hemoglobin, investigated the configurations of proteins and the effects of H-bonds. In 1949 he showed that sickle cell anemia was due to a mutation of a single amino acid in the hemoglobin molecule, the first correlation of a change in molecular structure with a genetic disease. Starting in 1951 he and colleagues published a series of papers describing the alpha helix structure of proteins; a paper published in the early 1950s with R.B. Corey on the structure of DNA played an important role in leading Francis Crick and James Watson to the double helix structure (Np 1962).

A further important achievement in the peptide field was that of Vincent Du Vigneaud (1901-1978, Np 1955), Cornell Medical School, who isolated and determined the structure of oxytocin **97**, a posterior pituitary gland hormone, for which a structure involving a disulfide bond was proposed. He synthesized oxytocin in 1953, thereby completing the first synthesis of a natural peptide hormone.



97. oxytocin
uterus-contracting and lactation-stimulating hormone of posterior pituatary gland
uterine contraction

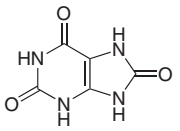
Progress in isolation, purification, crystallization methods, computers, and instrumentation, including cyclotrons, have made X-ray crystallography the major tool in structural biology in recent years. Numerous structures including those of ligand/receptor complexes are being published at an extremely rapid rate. Some of the past major achievements in protein structures are the following. Max Perutz (1914, Np 1962) and John Kendrew (1914-97, Np 1962), both at the Laboratory of Molecular Biology, Cambridge U., determined the structures of hemoglobin and myoglobin, respectively. William Lipscomb (1919-, Np 1976), Harvard U., who has trained many of the world's leaders in protein X-ray crystallography has been involved in the structure determination of many enzymes including carboxypeptidase A (1967); in 1965 he determined the structure of the anticancer bisindole alkaloid, vinblastine **98**. Folding of proteins, important but still enigmatic phenomenon, is attracting increasing attention. Christian Anfinsen (1916-1995, Np 1972), NIH, one of the pioneers in this area, showed that the amino acid residues in ribonuclease interact in an energetically most favorable manner to produce the unique 3D structure of the protein.



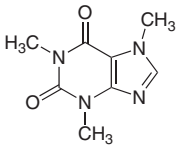
98. vinblastine
Vinca rosea
antineoplastic

Nucleic acid bases, RNA and DNA.

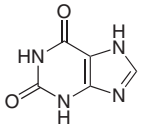
The "Fischer indole synthesis" was first performed in 1886 by Emil Fischer. During the period 1881-1914, he determined the structures of and synthesized uric acid **99**, caffeine **100**, xanthine **101**, guanine **102**, hypoxanthine **103**, adenine **104**, guanine **105**, and made theophylline or **106**-D-glucoside phosphoric acid, the first synthetic nucleotide. In 1903, he made 5,5-diethylbarbituric acid or Barbital **107**, Dorminal, Veronal, etc. (sedative), and in 1912, phenobarbital or Barbipil, Luminal, Phenobal, etc. (sedative). Many of his syntheses formed the basis of German industrial production of purine bases. In 1912 he showed that tannins are gallates of sugars such as maltose and glucose. Starting in 1899, he synthesized many of the thirteen α-amino acids known at that time, including the L- and D-forms, which were separated through fractional crystallization of their salts with optically active bases. He also developed a method for synthesizing fragments of proteins, namely peptides and made an 18 amino acid peptide. He lost his two sons in W.W.I, lost his wealth due to postwar inflation, believed he had terminal cancer (a misdiagnosis), and killed himself in July 1919. Fischer was a skilled experimentalist, so that even today, many of the reactions performed by him and his students are so delicately controlled that they are not easy to reproduce. As a result of his suffering by inhaling diethylmercury, and of the poisonous effect of phehylhydrazine, he was one of the first to design fume hoods. He was a superb teacher and was also influential in establishing the Kaiser Wilhelm Institute, which later became the Max Planck Institute. The number and quality of his accomplishments and contributions are hard to believe; he was truly a genius.



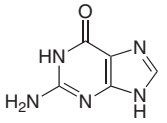
99. uric acid
discovered by Scheele and by Bergman in 1776



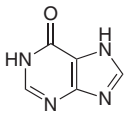
100. caffeine
tea, coffee, cola
CNS stimulant



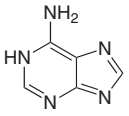
101. xanthine
animal organs, yeast, potatoes, tea



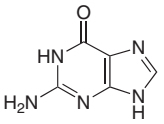
102. guanine
nucleic acid constituent



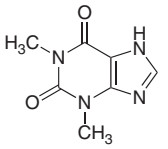
103. hypoxanthine
in animal body from nucleic acid breakdown



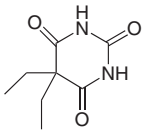
104. adenine
anaimal, plant tissues



105. guanine
animals, plants



106. theophylline
xanthine deriv.
diuretic, cardiac stimulant



107. barbital
Synth.: Fischer
sedative, hypnotic

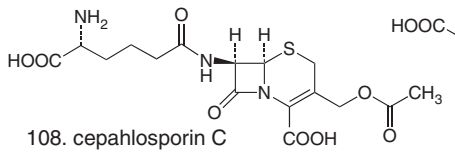
Alexander Todd (1907-1997, Np 1957) made critical contributions to the basic chemistry and synthesis of nucleotides. His early experience consisted of an extremely fruitful stay at Oxford in the Robinson group, where he completed the syntheses of many representative anthocyanins, and then at Edinburgh where he worked on the synthesis of vitamin B_1. He also prepared the hexacarboxylate of vitamin B_{12} (1954), which was used by D. Hodgkin's group for their X-ray elucidation of this vitamin (1956). M. Wiewiorowski (1918-), Inst. Bioorg. Chem., in Poznan, has headed a famous group in nucleic acid chemistry, and has produced colleagues distributed world-wide.

Antibiotics, Pigments, Marine Natural Products.

The concept of one microorganism killing another was introduced by Pasteur who coined the term antibiosis in 1877, but it was much later that this concept was realized in the form of an actual antibiotic. The bacteriologist Alexander Fleming (1881-1955, London U., Np 1945) noticed that an airborne mold, a Penicillium strain, contaminated cultures of *Staphylococci* left on the open bench and formed a transparent circle around its colony

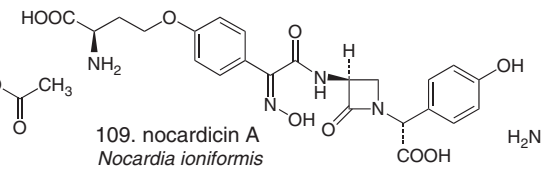
due to lysis of *Staphylococci*. He published these results in 1929. The discovery did not attract much interest but the work was continued by Fleming until it was taken up further at Oxford U. by pathologist Howard Florey (1898-1968, Np 1945) and biochemist Ernst Chain (1906-1979, Np 1945). The bioactivities of purified "penicillin", the first antibiotic, attracted serious interest in the early 1940s in the midst of W.W.II. A UK/USA team was formed during the war between academe/industry with Oxford U., Harvard U., ICI, Glaxo, Burroughs Welcome, Merck, Shell, Squibb and Pfizer as members. This project resulted in the large scale production of penicillin and determination of its structure as depicted in structure **28** (finally by X-ray, D. Hodgkin). John Sheehan (1915-1992) at MIT synthesized 6-aminopenicillanic acid in 1959, which opened the route for the synthesis of a number of analogs. Besides being the first antibiotic to be discovered, penicillin is also the first member of a large number of important antibiotics containing the β-lactam ring, e.g., cephalosporins **108**, carbapenems, monobactams, nocardicins **109**. The strained β-lactam ring of these antibiotics inactivates the transpeptidase by acylating its serine residue at the active site, thus preventing the enzyme from forming the link between the pentaglycine chain and the D-Ala-D-Ala peptide, the essential link in bacterial cell walls. The overuse of β-lactam antibiotics, which has given rise to the disturbing appearance of microbial resistant strains, is leading to active research in the design of synthetic β-lactam analogs to counteract these strains. The complex nature of the important penicillin biosynthesis is being elucidated through efforts combining genetic engineering, expression of biosynthetic genes as well as feeding of synthetic precursors, etc. by Jack Baldwin (1938-, Oxford U.), José Luengo (U. de León, Spain) and many other groups from industry and academe.

Shortly after the penicillin discovery, Selman Waksman (1888-1973, Rutgers U., Np 1952) discovered streptomycin **110**, the second antibiotic and the first active against the dreaded disease tuberculosis. The discovery and development of new antibiotics continued throughout the world at pharmaceutical companies in Europe, Japan and USA from soil and various odd sources: cephalosporin from sewage in Sardinia, cyclosporin **111** from Wisconsin and Norway soil which was carried back to Switzerland, avermectin **112** from the soil near a golf course in Shizuoka Prefecture. People involved in antibiotic discovery used to collect soil samples from various sources during their trips but this has now become severely restricted to protect a country's right to its soil. M.M. Shemyakin (1908-1970, Inst. Chem. Natural Products, Moscow) was a grand master of Russian natural products who worked on antibiotics, especially of the tetracycline class; he also worked on cyclic antibiotics composed of alternating sequences of amides and esters and coined the term depsipeptide for these in 1953. He died in 1970 of a sudden heart attack in the midst of the 7th IUPAC Natural Products Symposium held in Riga, Latvia, which he had organized. The Institute he headed was renamed the Shemyakin Institute.



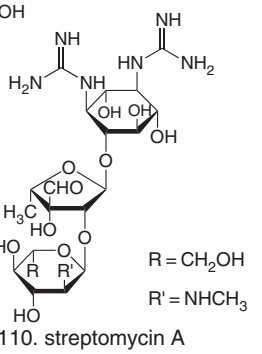
108. cepahlosporin C

Cephalosporium sp. together with pen. N from sea water near sewage outlet in Sardinia antibiotic



109. nocardicin A

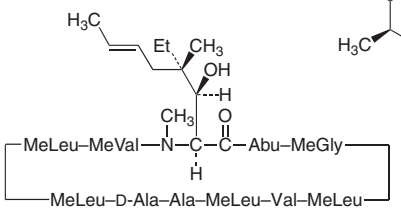
Nocardia ioniformis



110. streptomycin A

Streptmyces griseus

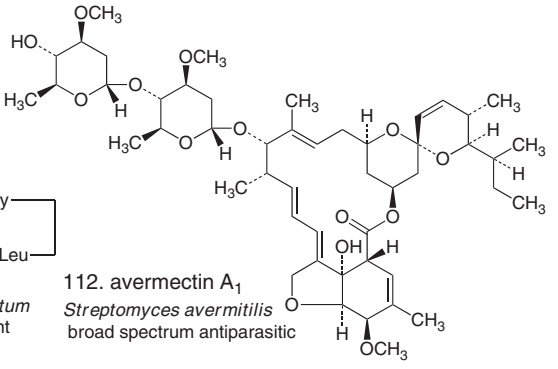
second antibiotic after penicillin against tuberculosis



111. cyclosporin A

Tolypocladium inflatum

immuno suppresant

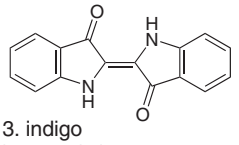


112. avermectin A_1

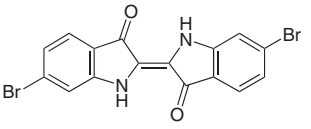
Streptomyces avermitilis

broad spectrum antiparasitic

Indigo **113**, an important vat dye known in ancient Asia, Egypt, Greece, Rome, Britain, and Peru, is probably the oldest known coloring material of plant origin, *Indigofera* and *Isatis*. The structure was determined in 1883 and a commercially feasible synthesis was performed in 1883 by Adolf von Baeyer (see above, 1835-1917, Np 1905), who founded the German Chemical Society in 1867 following the precedent of the Chemical Society of London. In 1872 Baeyer was appointed a professor at Strasbourg where E. Fischer was his student, and in 1875 he succeeded J. Liebig in Münich. Tyrian (or Phoenician) purple **114**, the dibromo derivative of indigo which is obtained from the purple snail *Murex bundaris*, was used as a royal emblem in connection with religious ceremonies because of its rarity; because of the availability of other cheaper dyes with similar color, it has no commercial value today. K. Venkataraman (1901-81, U. Bombay then National Chemical Lab.) who worked with R. Robinson on the synthesis of chromones in his early career, continued to study natural and synthetic coloring matters, including synthetic anthraquinone vat dyes, natural quinonoid pigments, etc. T.R. Seshadri (1900-75) is another Indian natural products chemist who worked mainly in natural pigments, dyes, drugs, insecticides, and especially in polyphenols. He also studied with Robinson, and with Pregl at Graz, and taught at Delhi U. Seshadri and Venkataraman had a huge impact on Indian chemistry.

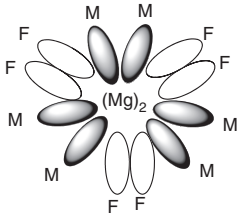


113. indigo
oldest known coloring matter
from plants; textile dye

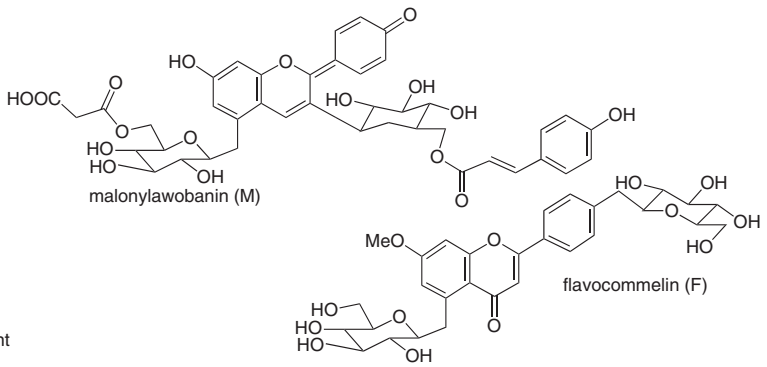


114. Tyrian purple
Murex bundaris (a snail)

After a 40 year involvement, Toshio Goto (1929-90) finally succeeded in solving the mysterious identity of commelinin, the deep-blue flower petal pigment of the *Commelina communis* isolated by Kozo Hayashi (1958) and protocyanin, isolated from the blue cornflower *Centaurea cyanus* by E. Bayer (1957). His group elucidated the remarkable structure **115** for commelinin in its entirety which consisted of six unstable anthocyanins, six flavones and two metals, a "metalloanthocyanin", MW 8846; complex stacking and hydrogen bonds were also involved. The structure of protocyanin is a similar supramolecule. Thus the pigmentation of petals turned out to be far more complex than the theories put forth by Willstätter (1913) and Robinson (1931). Goto suffered a fatal heart attack while inspecting the first X-ray structure of commelinin; commelinin represents a pinnacle of current natural products isolation and structure determination in terms of subtlety in isolation and complexity of structure. Goto solved many other challenging problems, including tetrodotoxin and luciferin (see below), and was a central figure in natural products research.



115. commelinin
Commelina communis
deep blue petal pigment

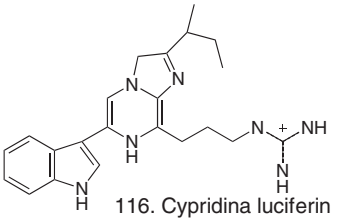


malonylawobanin (M)

flavocommelin (F)

The study of marine natural products is understandably far behind that of compounds of terrestrial origin due to the difficulty in collection and identification of marine organisms. On the other hand, it is an area which has great potentialities for new discoveries from every conceivable source. One pioneer in modern marine chemistry is Paul Scheuer (1915- , U. Hawai'i) who started his work with quinones of marine origin and since has characterized a very large number of bioactive compounds from mollusks and other sources. Luigi Minale (1936-1997, Napoli) started an active group working on marine natural products, concentrating mainly on complex saponins. He was a leading natural products chemist who died prematurely. A. Gonzalez Gonzalez (1917-) who headed the Organic Natural Products Institute at the U. of La Laguna, Tenerife, was the first to isolate and study polyhalogenated sesquiterpenoids from marine sources. His group has also carried out extensive studies on terrestrial terpenoids from the Canary Islands and South America. Carotenoids are widely distributed in nature and are of importance as food coloring material and as antioxidants (the detailed mechanisms of which still have to be worked out); new carotenoids continue to be discovered from marine sources, for example by the group of Synnove Liaaen-Jensen, Norwegian Inst. Tech.).

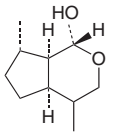
Yoshimasa Hirata (1915-), who started research at Nagoya U., is a champion in the isolation of nontrivial natural products. He characterized the bioluminescent luciferin **116** from the marine ostracod *Cypridina hilgendorfii* in 1966 (with his students, Toshio Goto, Yoshito Kishi and Osamu Shimomura); tetrodotoxin from the fugu fish in 1964 (with Goto and Kishi and coworkers); the structure was announced simultaneously by the group of Kyosuke Tsuda (1907- , tetrodotoxin, matrine) and Woodward; and the very complex palytoxin, $C_{129}H_{223}N_3O_{54}$ in 1981-87 (with Daisuke Uemura and Kishi. Richard E. Moore, U. Hawai'i, also announced the structure of palytoxin **117** independently. Jon Clardy (1943-, Cornell U.) has determined the X-ray structures of many unique marine natural products, including brevetoxin B **118** (1981), the first of the group of toxins with contiguous trans-fused ether rings constituting a stiff ladder-like skeleton. Maitotoxin **119**, MW 3422, $C_{164}H_{256}O_{68}S_2Na_2$ produced by the dinoflagellate *Gambierdiscus toxicus* is the largest and most toxic of the nonbiopolymeric toxins known; it has 32 alicyclic 6-8 membered ethereal rings and acyclic chains. Its isolation (1994) and complete structure determination was accomplished jointly by the groups of Takeshi Yasumoto (Tohoku U.), Kazuo Tachibana and Michio Murata (Tokyo U.) in 1996. Kishi, Harvard U., also deduced the full structure in 1996.



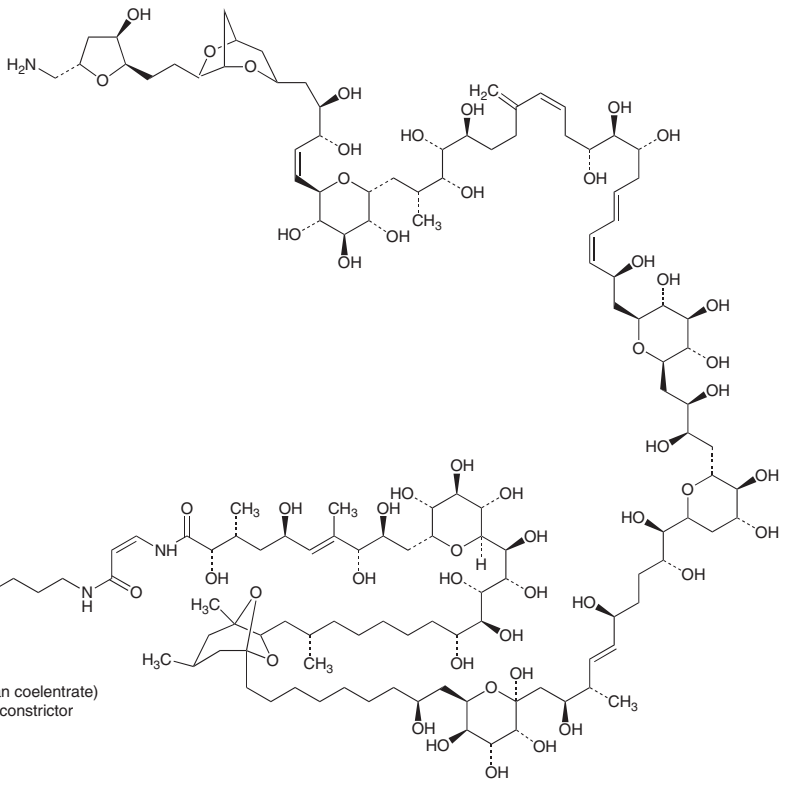
116. Cypridina luciferin
Cypridina hilgendorfii
First bioluminescent substance

The well-known excitatory agent for the cat family contained in the volatile oil of catnip *Nepeta cataria*, is the monoterpene nepetalactone **13**, isolated by S.M. McElvain (1943) and structure determined by Jerrold Meinwald (1954); cats, tigers and lions start purring and roll on their backs in response to this lactone. Takeo Sakan (1912-1993) investigated the series of monoterpenes neomatatabiols **120**, etc. from *Actinidia*, some of which are male lacewing attractants. As little as 1 femtogram of neomatatabiol attracts lacewings. The first insect pheromone to be isolated and characterized was bombykol, the sex attractant for the male silkworm, Bombyx mori (by Butenandt and coworkers, see above). Numerous pheromones have been isolated, characterized, synthesized and are playing central roles in insect control and in chemical ecology. The group at Cornell U. have long been active in this field: Tom Eisner (1929-,behavior), Jerrold Meinwald (1927-, chemistry), Wendell Roeloff (1938-, electrophysiology, chemistry). Since the available sample is usually minuscule, full structure determination of a pheromone often requires total synthesis; Kenji Mori (1935-, Tokyo U.) has been particularly active in this field. Progress in the techniques for handling volatile compounds, including

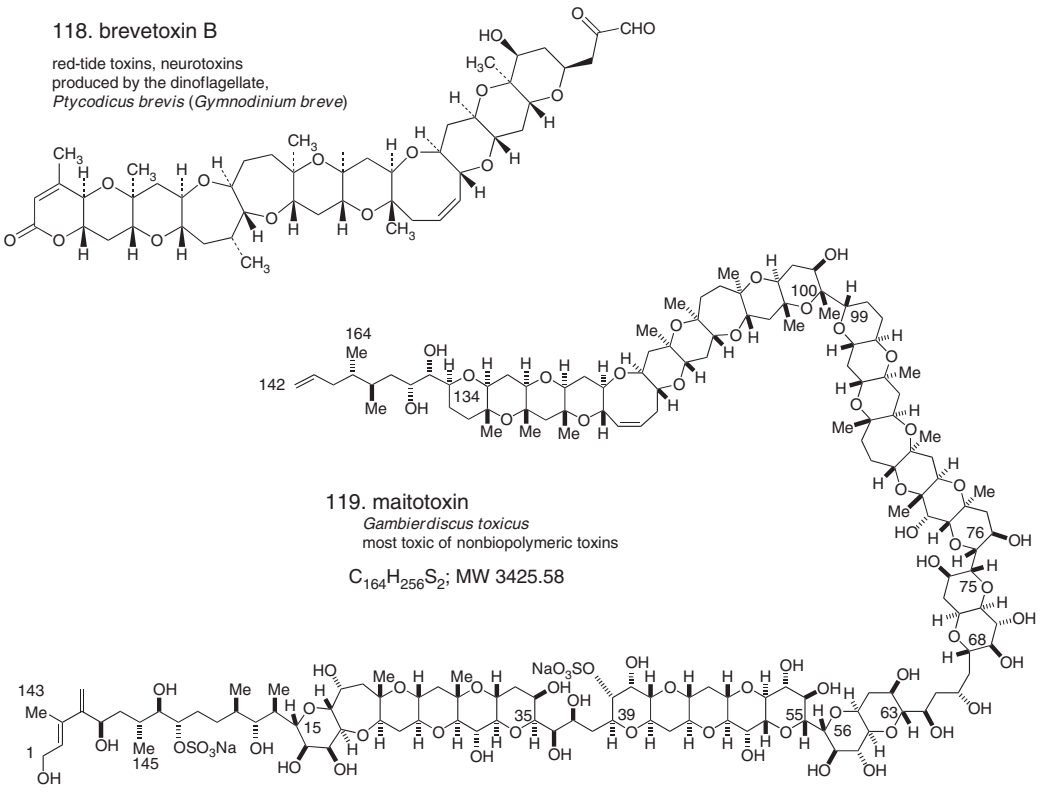
collection, isolation, GC/MS, etc. has started to disclose the extreme complexity of chemical ecology which plays an important role in the lives of all living organisms. In this context, natural products chemistry will be play an increasingly important role in our grasp of the significance of biodiversity.



120. neomatatabiol
Actidinia polygama
male lacewing attractant, 1ng level



117. palytoxin
Palythoa toxica (Hawaiian coelentrate)
highly toxic, intense vasoconstrictor



Synthesis

Synthesis has been mentioned often in the preceding sections of this essay. In the following, synthetic methods of more general nature are described. The Grignard reaction of Victor Grignard (1871-1935, Np 1912) and then the Diels-Alder reaction by Otto Diels (1876-1954, Np 1950) and Kurt Alder (1902-1956, Np 1950) are extremely versatile reactions. The Diels-Alder reaction can account for the biosynthesis of several natural products with complex structures, and now an enzyme, a Diels-Alderase involved in biosynthesis has been isolated by Akitami Ichihara, Hokkaido U. (1997).

The hydroboration reactions of Herbert Brown (1912-, Purdue U., Np 1979) and the Wittig reactions of Georg Wittig (1897-1987, Np 1979) are extremely versatile synthetic reactions. William S. Johnson (1913-1995, U. Wisconsin, Stanford U.) developed efficient methods for the cyclization of acyclic poly-olefinic compounds for the synthesis of corticoid and other steroids, while Gilbert Stork (1921- , Columbia U) introduced enamine alkylation, regiospecific enolate formation from enones and their kinetic trapping (called "three component coupling" in some cases), and radical cyclization in regio- and stereospecific constructions. Elias J. Corey (1928-, Harvard U., Np 1990) introduced the concept of retrosynthetic analysis and developed many key synthetic reactions and reagents during his synthesis of bioactive compounds, including prostaglandins and gingkolides. A recent development is the ever-expanding supramolecular chemistry stemming from 1967 studies on crown ethers by Charles Pedersen (1904-1989), 1968 studies on cryptates by Jean-Marie Lehn (1939-), and 1973 studies on host-guest chemistry by Donald Cram (1919-); they shared the chemistry Nobel prize in 1987.

Natural products studies in Japan.

Since the background of natural products study in Japan is quite different from that in other countries, a brief history is given here. Natural products is one of the strongest areas of chemical research in Japan with probably the world's largest number of chemists pursuing structural studies; these are joined by a healthy number of synthetic and bioorganic chemists. An important Symposium on Natural Products was held in 1957 in Nagoya as a joint event between the faculties of science, pharmacy and agriculture. This was the beginning of a series of annual symposia held in various cities, which has grown into a three- day event with about 50 talks and numerous posters; practically all achievements in this area are presented at this symposium. Japan adopted the early 20th century German or European academic sytem where continuity of research can be assured through a permanent staff in addition to the professor, a system which is suited for natural products research which involves isolation, assay, as well as structure determination, all steps requiring delicate skills and much expertise.

The history of Japanese chemistry is short because the country was closed to the outside world up to 1868. This is when the Tokugawa shogunate which had ruled Japan for 264 years was overthrown and the Meiji era (1868-1912) began. Two of the first Japanese organic chemists sent abroad were Shokei Shibata and Nagayoshi Nagai, who joined the laboratory of A. W. von Hoffmann in Berlin. Upon return to Japan, Shibata (Chinese herbs) started a line of distinguished chemists, Keita and Yuji Shibata (flavones) and Shoji Shibata (1915-, lichens, fungal bisanthraquinonoid pigments, ginsenosides); Nagai returned to Tokyo Science University in 1884, studied ephedrine, and left a big mark in the embryonic era of organic chemistry. Modern natural products chemistry took a real start when three extraordinary organic chemists returned from Europe in the 1910s and started research/teaching at their respective faculties:

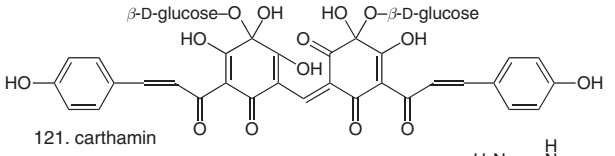
Riko Majima, 1874-1962, C.D. Harries (Kiel U); R. Willstätter (Zürich): Faculty of science, Tohoku U.; urushiol, the catecholic mixture of poison ivy irritant.

Yasuhiko Asahina, 1881-1975, R. Willstätter: Faculty of pharmacy, Tokyo U.; lichens and Chinese herb.

Umetaro Suzuki, 1874-1943, E. Fischer: Faculty of agriculture, Tokyo U.; vitamin B_1(thiamine).

Because these three pioneers started research in three different faculties (i.e., science, pharmacy, agriculture), and because little interfaculty personnel exchange occurred in subsequent years, natural products chemistry in Japan was pursued independently within these three academic domains; the situation has changed now. The three pioneers started lines of first class successors, but the establishment of a strong infrastructure takes many years, and it is only after the mid-1960s that the general level of science became comparable to that in the rest of the world; the 3rd IUPAC Symposium on the Chemistry of Natural Products, presided over by Munio Kotake (1894-1976, bufotoxins, see below), held in 1964 in Kyoto, was a clear turning point in Japan's role in this area.

Some of the outstanding Japanese chemists not already quoted are the following. Shibasaburo Kitazato (1852-1931), worked with Robert Koch (Np 1905, tuberculosis) and von Behring, antitoxins of diphtheria and tetanus which opened the new field of serology, isolation of microorganism causing dysentery, founder of Kitazato Inst.; Chika Kuroda (1884-1968), first female Ph.D., structure of the complex carthamin **121**, important dye in safflower (1930) which was revised in 1979 by Obara et al., although the absolute configuration is still unknown (1998); Munio Kotake (1894-1976), bufotoxins **122**, tryptophan metabolites, nupharidine **123**; Harusada Suginome (1892-1972), aconite alkaloids; Teijiro Yabuta (1888-1977), kojic acid **124**, gibberrelins **125**; Eiji Ochiai (1898-1974), aconite alkaloids; Toshio Hoshino (1899-1979), abrine and other alkaloids; Yusuke Sumiki (1901-74), gibberrelins; Sankichi Takei (1896-1982) rotenone **126**; Shiro Akabori (1900-1992), peptides, C-terminal hydrazinolysis of amino acid ; Hamao Umezawa (1914-86), kanamycin **127**, bleomycin **128**, numerous antibiotics; Shojiro Uyeo (1909-88), lycorine **129**; Tsunematsu Takemoto (1913-89), inokosterone **130**, domoic acid **131**, kainic acid **132**, quisqualic acid **133**; Tomihide Shimizu (1889-58), bile acids; Kenichi Takeda (1907-1991), Chinese herbs, sesquiterpenes; Yoshio Ban (1921-94), alkaloid synthesis; Wataru Nagata (1922-1993), stereocontrolled hydrocyanation.

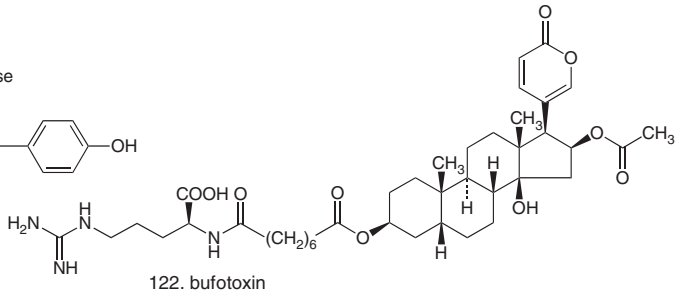


121. carthamin

Carthamus tinctorius (safflower, natura; red)

dye

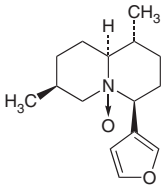
absol. config. unknown



122. bufotoxin

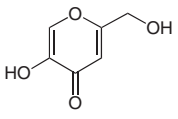
toxin of European toad *Bufo vulgaris*

isoln., structure: Wieland, Alles 1922



123. nupharidine

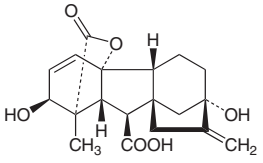
rhizome of *Nuphar*



124. kojic acid

Aspergillus oryzae, others.

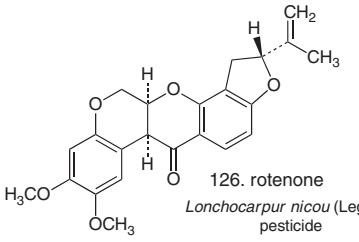
antibiotic, flavor enhancing additive



125. gibberellin A_3

Gibberella fujikuroi

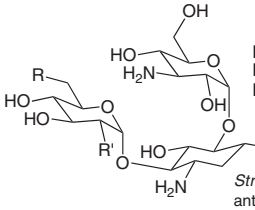
plant hormone, growth promoter



126. rotenone

Lonchocarpur nicou (Legum.)

pesticide

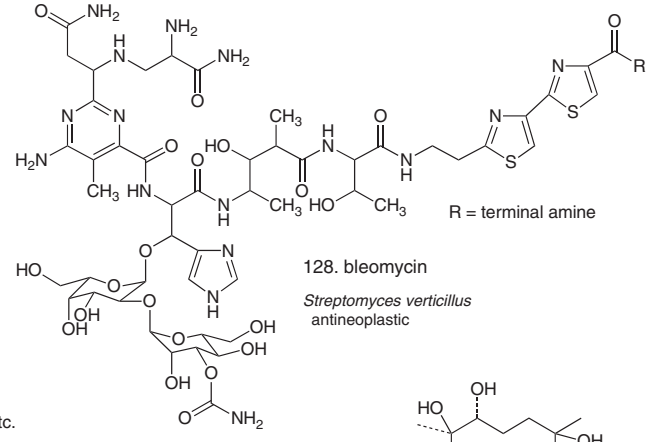


	R	R'
Kanamycin A	NH_2	OH
Kanamycin B	NH_2	NH_2
Kanamycin C	OH	NH_2

127. kanamycin

Streptomyces kanamyceticus

antibacterial

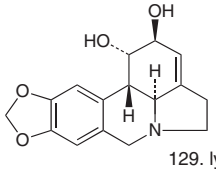


R = terminal amine

128. bleomycin

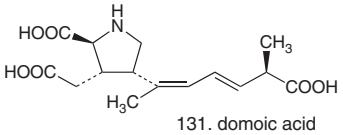
Streptomyces verticillus

antineoplastic



129. lycorine

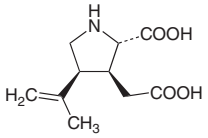
bulbs of *Narcissus pseudonarcissus*, etc.



131. domoic acid

red algae *Chondra armata*

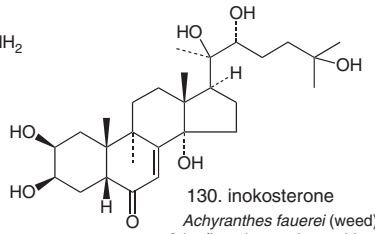
excitatory amino acid



132. kainic acid

red algae *Diginea simplex*, etc.

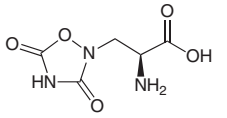
anthelmintic, used in neurobiol.



130. inokosterone

Achyranthes fauerei (weed)

one of the first phytoecdysteroids



133. quisqualic acid

seeds of *Quisqualis chinensis*

anthelmintic, used in neurobiol.

132/133 are used for identifying specific subset of excitatory amino acid receptors, i.e., kainate receptors, quisqualate receptors.

Current and future trends in natural products chemistry

Spectroscopy and X-ray crystallography has totally changed the process of structure determination, which used to generate the excitement of solving a mystery. The first introduction of spectroscopy to the general organic community was Woodward's 1942-1943 empirical rules for estimating the UV maxima of dienes, trienes, and

enones, which were extended by Fieser (1959). However, Butenandt had used UV for correctly determining the structures of the sex hormones as early as the early 1930s, while Karrer and Kuhn also used UV very early in their structural studies of the carotenoids. The Beckman DU instruments were an important factor which made UV spectroscopy a common tool for organic chemists and biochemists. With the availability of commercial instruments in 1950, IR spectroscopy became the next physical tool, making the 1950 Colthup IR correlation chart and the 1954 Bellamy monograph indispensable. The IR fingerprint region was analyzed in detail in attempts to gain as much structural information as possible from the molecular stretching and bending vibrations. Introduction of NMR spectroscopy into organic chemistry, first for protons and then for carbons, has totally changed the picture of structure determination, so that now IR is used much less frequently; however, in biopolymer studies, the techniques of difference FTIR and resonance Raman spectroscopy are indispensable.

The dramatic and rapid advancements in mass spectrometry are now drastically changing the protocol of bio-macromolecular structural studies performed in biochemistry and molecular biology. Herbert Hauptman (mathematician, 1917-, Med. Foundation, Buffalo, Np 1985) and Jerome Karle (1918-, US Naval Research Laboratory, Washington, DC, Np 1985) developed direct methods for the determination of crystal structures devoid of disproportionately heavy atoms. The direct method together with modern computers revolutionized the X-ray analysis of molecular structures, which has become routine for crystalline compounds, large as well as small. Fred McLafferty (1923, Cornell U.) and Klaus Biemann (1926-, MIT) have made important contributions in the development of organic and bioorganic mass spectrometry. The recent cyclotron-based facilities for crystallographic biology studies has led to further dramatic advances enabling some protein structures to be determined in a single day, while cryoscopic electron micrography developed in 1975 by Richard Henderson and Nigel Unwin has also become a powerful tool for 3D structural determinations of membrane proteins such as bacteriorhodopsin (25 kd) and the nicotinic acetylcholine receptor (270 kd).

Circular dichroism, which was used by French scientists Jean B. Biot (1774-1862) and Aimé Cotton during the last century "deteriorated" into monochromatic measurements at 589 nm after R.W. Bunsen (1811-1899, Heidelberg) introduced the Bunsen burner into the laboratory which readily emitted a 589 nm light characteristic of sodium. The 589 nm $[\alpha]_D$ values, remote from most chromophoric maxima, simply represent the summation of the low intensity readings of the decreasing end of multiple Cotton effects. It is therefore very difficult or impossible to deduce structural information from $[\alpha]_D$ readings. Chiroptical spectroscopy was re-introduced to organic chemistry in the 1950s by C. Djerassi at Wayne State U. (then Stanford U.) as optical rotatory dispersion (ORD) and by L. Velluz and M. Legrand at Roussel-Uclaf as circular dichroism. Günther Snatzke (1928-92, Bonn then Ruhr U. Bochum) was a major force in developing the theory and application of organic chiroptical spectroscopy. He investigated the chiroptical properties of a wide variety of natural products, including constituents of indigenous plants collected throughout the world, and established semiempirical sector rules for absolute configurational studies. He also established close collaborations with scientists of the former Eastern bloc countries and had a major impact in increasing the interest in CD there.

Chiroptical spectroscopy, nevertheless, remains one of the most underutilized physical measurements. Most organic chemists regard CD (more popular than ORD because interpretation is usually less ambiguous) simply as a tool for assigning absolute configurations, and since there are only two possibilities in absolute configurations, CD is apparently regarded not as crucial compared to other spectroscopic methods. Moreover, many of the CD correlations with absolute configuration are empirical. For such reasons, chiroptical spectroscopy, with its immense potentialities, is grossly underused. However, CD curves can now be calculated nonempirically. Moreover, through-space coupling between the electric transition moments of two or more chromophores gives rise to intense Cotton effects split into opposite signs, i.e., exciton coupled CD; fluorescence detected CD further enhances the sensitivity by 50 to 100-fold. This leads to a highly versatile nonempirical microscale solution method for determining absolute configurations, etc.

With the rapid advances in spectroscopy and isolation techniques, most structure determinations in natural products chemistry have become quite routine, shifting the trend gradually towards activity-monitored isolation and structural studies of biologically active principles available only in microgram or submicrogram

quantities. This in turn has made it possible in recent years for organic chemists to direct their attention towards clarifying the mechanistic and structural aspects of the ligand / biopolymeric receptor interactions on a more well-defined molecular structural basis. A decade ago, it was inconceivable and impossible to perform such studies.

Why does sugar taste sweet? This is an extremely challenging problem which at present cannot be answered even with major multidisciplinary efforts. Structural characterization of sweet compounds and elucidation of the amino acid sequences in the receptors are only the starting point. We are confronted with a long list of problems such as cloning of the receptors to produce them in sufficient quantities to investigate the physical fit between the active factor (sugar) and receptor by biophysical methods, and the time-resolved change in this physical contact and subsequent activation of G-protein and enzymes. This would then be followed by neurophysiological and ultimately physiological and psychological studies of sensation. How do the hundreds of taste receptors differ in their structures and their physical contact with molecules, and how do we differentiate the various taste sensations? The same applies to vision and to olfactory processes. What are the functions of the numerous glutamate receptor subtypes in our brain? We are at the starting point of a new field which is filled with exciting problems.

Familiarity with molecular biology is becoming essential for natural products chemists to plan research directed towards an understanding of natural products biosynthesis, mechanisms of bioactivity triggered by ligand-receptor interactions, etc. Numerous genes encoding enzymes have been cloned and expressed by the cDNA and/or genomic DNA-polymerase chain reaction protocols. This then leads to the possible production of new molecules by gene shuffling and recombinant biosynthetic techniques. Monoclonal catalytic antibodies using haptens possessing a structure similar to a high-energy intermediate of a proposed reaction is also contributing to the elucidation of biochemical mechanisms and the design of efficient syntheses. The technique of photoaffinity labeling, brilliantly invented by Frank Westheimer (1912- , Harvard U.), assisted especially by advances in mass spectrometry, will clearly be playing an increasingly important role in studies of ligand/receptor interactions including enzyme/substrate reactions. The combined and sophisticated use of various spectroscopic means, including difference spectroscopy and fast time-resolved spectroscopy, will also become increasingly central in future studies of ligand/receptor studies.

Organic chemists, especially those involved in structural studies have the techniques, imagination, and knowledge to use these approaches. But it is difficult for organic chemists to identify an exciting and worthwhile topic. In contrast, the biochemists, biologists, and medical doctors are daily facing exciting life-related phenomena, frequently without realizing that the phenomena could be understood or at least clarified on a chemical basis. Broad individual expertise and knowledge coupled with multidisciplinary research collaboration thus becomes essential to investigate many of the more important future targets successfully. This approach may be termed “dynamic”, as opposed to a “static” approach, exemplified by isolation and structure determination of a single natural product. Fortunately for scientists, nature is extremely complex and hence all the more challenging. Natural products chemistry will be playing an absolutely indispensable role for the future. Conservation of the alarming number of disappearing species, utilization of biodiversity, and understanding of the intricacies of biodiversity are further difficult but urgent problems confronting us.

That natural medicines are attracting renewed attention is encouraging from both practical and scientific viewpoints; their efficacy has often been proven over the centuries. However, to understand the mode of action of folk herbs and related products from nature, is even more complex than mechanistic clarification of a single bioactive factor. This is because unfractionated or partly fractionated extracts are used, often containing mixtures of materials, and in many cases synergism is most likely playing an important role. Clarification of the active constituents and their modes of action will be difficult. This is nevertheless a worthwhile subject for serious investigations.

Dedicated to Sir Derek Barton who planned this series with amazing insight, but passed away just before its completion. It is a pity that he was unable to write this article as originally planned, since he would have had a masterful overview of the content he wanted, based on his vast experience. I have tried to fulfill his task, but this article cannot do justice to his original intention.

Acknowledgment

I am grateful to current group members for letting me to take the time off to undertake this assignment unprepared. I am grateful to Drs. Nina Berova, Reimar Bruening, Jerrold Meinwald, Yoko Naya, Tetsuo Shiba and many other friends for suggestions.

References

A 100 Year History of Japanese Chemistry, ed. Chem. Soc. Japan, Tokyo Kagaku Dojin (1978).
Advanced Organic Chemistry, Louis F. Fieser & Mary Fieser, Reinhold Book Corporation, New York (1961).
Britannica Online, 1994-1998.
Bulletin of the Oriental Healing Arts Inst. U.S.A., Los Angeles, Vol 5, No. 7 (1980).
Murder, Magic and Medicine, John Mann, Oxford Univ. Press, New York (1992).
Natural Products Related to Phenanthrene, Louis F. Fieser & Mary Fieser, Reinhold Publishing Co. (1949).
Organic Molecules in Action, Murray Goodman, Frank Morehouse, Gordon & Breach Science Publishers, New York (1973).
Serendipity, Accidental Discoveries in Science, Royston M. Roberts, John Wiley & Sons, Inc., New York (1989).
The History of Organic chemistry in the United States, 1875-1955, Dean S. Tarbell, Tracey Tarbell, Folio Publishers, Nashville, Tennessee (1986).
The Origins of Intermediary Metabolism at Columbia College of Physicians and Surgeons (P & S), Konrad Bloch, The FASEB Journal, 10, 802-805 (1996).