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Leopold Ruzicka – From the Isoprene Rule to the Question of Life's Origin

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This is an English version of the article «Leopold Ruzicka: Von der Isoprenregel zur Frage nach dem Ursprung des Lebens» that appeared in the Leopold Ruzicka Centenary issue of Rad, the journal of the Jugoslavian Academy of Sciences and Arts, Zagreb (Rad, Jugoslav. akad. znan. umjet. kem. [443] 7, 21–68 (1989)). The German text was translated by Professor Dr. Eli Loewenthal, Department of Chemistry, Technion Haifa. Permission for publishing an English version was provided by Professor K. Balenović, Zagreb.

> *«In his later years he (Ruzicka) became greatly interested in aspects of evolution and of the origin of life which in his view endorsed his general philosophy, as expressed in many articles and books which he had collected and in the comments he made on this topic.»*
> (From *L. Ruzicka, Biographical Memoirs of Fellows of the Royal Society,* by V. Prelog and O. Jeger[1])

This short quotation hints at an underlying theme in *Ruzicka*'s scientific work. Barely perceptible as an undertone in some of his early publications, it later developed into a full-blown motif, if not basic motivation, after his formal retirement. We find this borne out by a wealth of material left after his death: in the form of hand-written notes and comments, marginal observations scribbled in books and other publications, and in addition, manuscript drafts of lectures, mostly left unpublished, under headings such as «Biosynthese heute und während der biologischen und chemischen Evolution»[2], or «Biochemie und Weltbild»[3]. The overall impression is that of an intense preoccupation on his part.

All of us who were fortunate to have been in close personal and professional contact with him in those days can well remember the single-mindedness with which he started to pursue the biochemical literature. It was as if at long last there came to the surface that innate driving force that was behind his work on terpenes – its biological significance, that aspect of chemistry which to him was of primary import in what he considered the order of things in the universe.

> *«Die wunderbare Harmonie, die zwischen den biochemischen Reaktionen in der lebenden Zelle herrscht, wäre eines Gottes würdig»*

– is an authentic remark made by *Ruzicka* at that time.

There is an inner connection between *Ruzicka*'s experimental work and his later inclination towards biochemistry that is discernible already in his early investigations on sesquiterpenes. They were not merely determination of constitution but the vehicle for recognizing the importance of a unifying principle in structure determination – the «Isoprene Rule», and ultimately for first indications of the significance of that rule for the recognition of the path by which nature constructs these molecules. In his 1933 address to the Zürcher Naturforschende Gesellschaft[4] he referred to the role of the chemistry of organic natural products as the way by which the natural products chemist could gain an indirect insight into the chemical processes of nature. And finally, in the 1950's, there was that enormous enthusiasm with which *Ruzicka*, soon to retire, regarded the structural and mechanistic connections between the sesqui-, di- and triterpenes, which were to find their formulation in the «Biogenetic Isoprene Rule»[5,6]. That was to be the culmination of his life's work and of the work of his school, which would unlock the portal between structure determination of terpenes and biochemistry.

Ruzicka's life and scientific work have already been summarized in the above-mentioned Biographical Memoir by *Prelog* and *Jeger*. In addition, the occasion of *Ruzicka*'s 100th birthday has been marked by publications dealing with specific aspects of his life's work[7], such as an essay by *Günther Ohloff* on his accomplishments as an inventor[7a].

The object of the present article is to depict *Ruzicka* as the – so to say – romantic scientist, shrewd realist though he was in his research, the one who never missed an opportunity to put his work on structure determination into the context of a «Chemistry of Life»[8], the one who as a boy had wanted to become a priest[9], who had then read *Häckel*'s «Welträtsel» in his student days[10], and who, though eventually fulfilled with the success of his scientific achievements, never lost the ability of being amazed at life's mysteries. These lines are motivated by the author's reminiscences of collaboration with *Ruzicka* after the doctorate and of *Ruzicka*'s influence on his own academic career. They are meant not only to express the author's opinions but above all to provide a forum for *Ruzicka*'s views, as formulated in published and unpublished lectures, notes and comments, and as shown by reproduction in his own characteristic handwriting.

It would be naive and misleading, however, to attempt to find a direct connection between aspects of *Ruzicka*'s personality as described above and his choice of scientific problems to tackle. In that he was first and foremost a solid and down-to-earth realist, with a healthy dose of opportunism when it was appropriate. In his own words (a Belgrade lecture towards the end of his career[11]):

> *«In all my work, there was at the beginning a common objective – to elucidate the structures of natural products which were of industrial importance ...»* (see Fig. 1).

In the sequel we shall see just how accurately that described the beginning of his independent scientific work.

In the Beginning

In 1916, after nearly six years as research assistant to *Hermann Staudinger*, his former mentor, *Ruzicka* felt that it was time for his «Habilitation» and the right to em-

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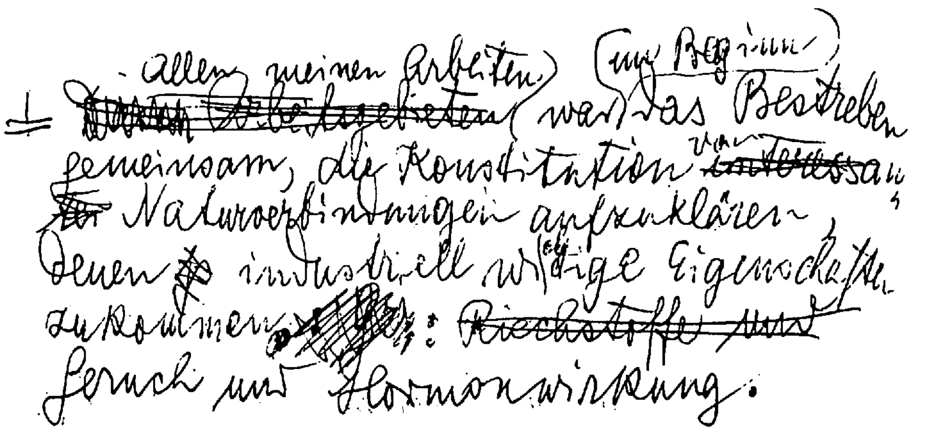
bark on his own independent scientific career.

«Toward the end of 5½ years of mainly synthetic work on the pyrethrins I had come to the firm conclusion that we were barking up the wrong tree» [12].

Since he now had to find financial support of his own, he decided to start on a synthesis of irone, an olfactory principle isolated in 1893 by *Tiemann* and *Krüger* [13] from iris root. To this end, and with *Staudinger*'s help, he arrived at an agreement of collaboration with Haarmann and Reimer in Holzminden, the oldest established perfumery company in Germany[14]. On the basis of the (wrong) empirical formula $C_{13}H_{20}O$ for irone, *Tiemann* and *Krüger* had effected a condensation reaction between citral and acetone ($C_{10}H_{16}O + C_3H_6O = C_{13}H_{20}O + H_2O$!), thus arriving at their well-known discovery of «ionone»[13], an artificial «essence of violets» whose subsequent manufacture by the Holzminden company was a great industrial success. On the basis of the supposed constitution of this «ionone» *Tiemann* had then suggested one for irone which was simply that of a double bond isomer of «ionone» (Fig. 2)[1*]. In his retrospective publication «Rolle der Riechstoffe in meinem chemischen Lebenswerk»[12] *Ruzicka* made the following comments:

«As I had no money of my own and could not retain my position of assistant I was glad to win the support of this company. However, they then informed me that the structure suggested for irone was in error since catalytic hydrogenation gave only a dihydro compound and therefore it would have to be bicyclic. I then immediately changed my plans and decided to embark on the synthesis of a bicyclic compound of ionone type, starting from the bicyclic terpene ketone fenchone [2*]*. For the latter Semmler had suggested a formula with three methyl groups situated as in the ionones and irone. On the other hand Wallach, in his 1910 Nobel lecture, had defined all monoterpenes as derivatives of p-cymene, thus rejecting Semmler's formula as being inconsistent with that definition* [3*]*. Since I was inclined to agree with Semmler I then decided to put an end to this controversy through a total synthesis.»*



1 – ~~[illegible]~~ Allen meinen Arbeiten (am Beginn) war das Bestreben
gemeinsam, die Konstitution von ~~interessan~~
~~für~~ Naturverbindungen aufzuklären,
denen industriell wichtige Eigenschaften
zukommen ~~[illegible]~~: ~~Riechstoffe und~~
Geruch und Hormonwirkung.

Fig. 1. From Ruzicka's manuscript for [11].

Nothing came out of that irone project, nor out of subsequent hectic pursuit of other supposedly irone-like compounds[14]. What did result, in 1917, was *Ruzicka*'s first independent publication entitled «The Total Synthesis of Fenchone»[19] which finally and conclusively confirmed the *Semmler* structure.

Again and again in his later years, *Ruzicka* returned in one context or another to that odoriferous constituent of the iris root which marked the onset of his career – a for him troublesome affair de cœur fittingly characterized as a «comedy of errors» in the Prelog-Jeger Biographical Memoir[1]. This continued well into the 1940's.

Olfactory substances played a decisive role in *Ruzicka*'s career, as he himself stressed at the age of 84[12], and it is of some

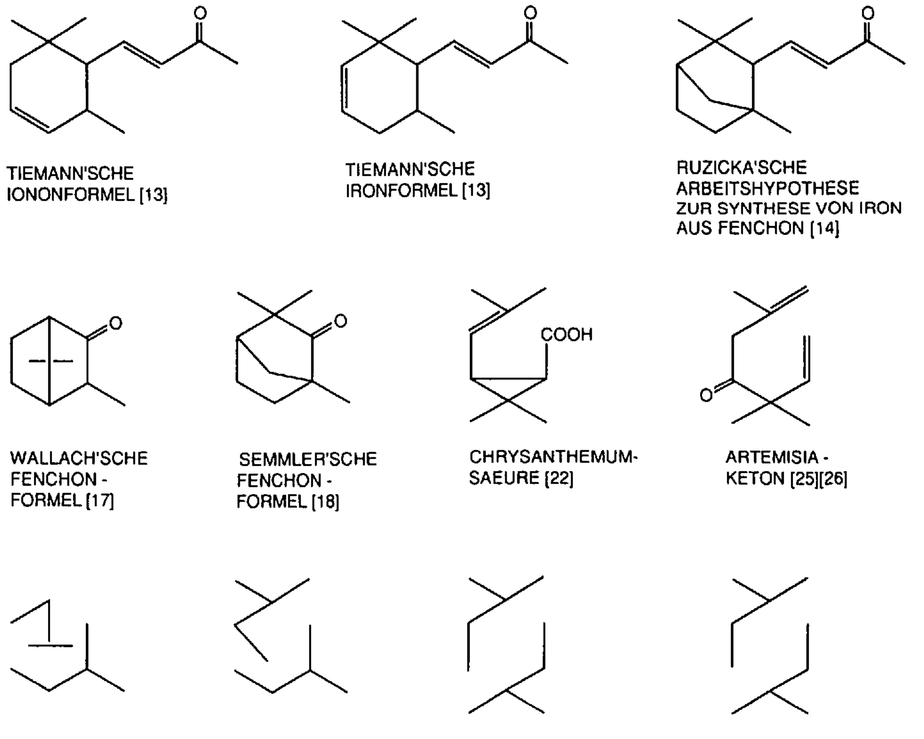


Fig. 2. Constitutional formulae which played an important role at the beginning of Ruzicka's scientific career.

[1*] Ironically enough, at the time of that historically important discovery, not only was there a mistake in the empirical formula ascribed to irone (corrected only 40 years later to $C_{14}H_{22}O$ by *Ruzicka, Seidel,* and *Schinz* [15]), but also in the constitutional formula for citral (both carbon-carbon double bonds conjugated with the aldehyde group). As a result of the latter, the constitutional formulae of the primary condensation product pseudoionone and of «ionone» were also incorrect (cf. Fig. 2). The discovery of «ionone» by its odour on treatment of the odoriferously uninteresting pseudoionone with sulfuric acid, represents in retrospect the discovery of the acid-catalyzed polyene cyclization, a reaction whose true nature was recognized only much later and which is probably the most important type of structural change encountered in the chemistry and biochemistry of the cyclic terpenes.

[2*] *«That is where we wanted to hang on an aldehyde group, condense the product with acetone and see whether we get something that smells like irone»* (from a recording of *Ruzicka*'s "Chemical Autobiography" lecture, Swiss Chemical Society, Basel, 13 September 1957).

[3*] This is where *Ruzicka* (at the age of 84!) is mistaken: there is no mention of fenchone in *Wallach*'s Nobel lecture[16]. In his book "Terpenes and Camphor"[17], pp. 531–539, *Wallach* justifies his own structure for fenchone by the fact of its conversion into *meta*-cymene (Fig. 2), admits that the Semmler structure[18] might be correct, but suggests that for the time being there is not enough experimental evidence to justify «abandoning the old structure». *Ruzicka*'s own paper on fenchone[19] makes no mention of *Wallach*'s *para*-cymene rule.

interest to reflect on this. There is no indication in his doctoral dissertation («On Phenyl Methyl Ketene»[20]); but quite probably subsequent work with *Staudinger* during the years 1911 to 1916 on the insecticidal components of Compositae had something to do with his predilection-to-be. In the publication of that work (on «Dalmatian Insect powder» from the ground-up petals of *Chrysanthemum cinerariifolium* Bocc.[21]) we find the following observation:

> *«For further purification we distilled the crude oil in a high vacuum. We obtained a forerun of a volatile material which had the characteristic odour of the insect powder but none of its insecticidal activity»* [21].

The active components of that powder, the pyrethrins, turned out to be alicyclic compounds. *Ruzicka* isolated and characterized a product $C_{10}H_{16}O_2$[22] (Fig. 2) which he named chrysanthemic acid; its ozonolysis led to caronic acid (total synthesis by *Perkin* and *Thorpe*[23] in 1899), a degradation product of the monoterpene carvone. It can have been no later than the time of this idenfication of caronic acid (1912) that *Ruzicka* became acquainted with the monoterpene chemical literature. An unpublished manuscript of a *Ruzicka* lecture of an autobiographical nature states:

> *«For me the pyrethrins were fate; and all my life I remained devoted to alicyclic compounds.»*

Was it *Tiemann* and *Krüger* who had a decisive influence on him? It is conceivable that the remarkable success story of these two pioneers, in arriving at the discovery of a commercially successful synthetic perfume through shortcutting a structure determination of a natural product by synthesis (see [1*]), made such a lasting impression on him, particularly when taking into account his background, personality, and material circumstances at that time. Indeed, in retrospect the story of ionone became a prototype for *Ruzicka*'s later successes: the syntheses of nerolidol and farnesol, the structure determination of civet and musk constituents leading to the syntheses of large-ring ketones, the proof of constitution of androsterone through a sensationally simple partial syntheses of this, and of the other male hormone testosterone. All of these exhibit the stamp of the same grand pattern: the choice of a natural product objective of both scientific and industrial significance, a daring working hypothesis on its structure, followed by a bold and straight-to-the-point synthesis (low yields notwithstanding!); and at the end universal recognition and financial reward. The *Ruzicka* of the isoprene rule was above all a man of concrete and affirmative action, of «proof of structure by synthesis»; he was a great expert neither on synthetic methodology nor on complex analytical deduction. The latter, so contrary to the accepted picture of *Ruzicka*, is attested to by his long drawn out and mostly abortive work on the structures of irone, cedrene, caryophyllene, and elemol.

It was quite in character for him to have been the first to use systematically the direct and harsh method of dehydrogenation in structure elucidation of polyterpenes. He was not one for tedious and protracted routes of oxidative degradation, nor for their meticulous and painstaking rationalization. This is not refuted by the work on elucidating the structure of lanosterol – the credit for that spectacular achievement in the field of solving chemical structures by classical degradation methods is in fact mainly due to *Oskar Jeger*, *Ruzicka*'s former student and later head of his triterpene research group. Speculation and risk-taking were decidedly part of *Ruzicka*'s character. His good fortune in always succeeding was due to his innate integrity, his enormous industriousness, and his unconditional belief in the experiment.

Toward the Extension of the Isoprene Rule

Both fenchone and chrysanthemic acid (see Fig. 2) are C_{10}-compounds whose structures as known at the time decidedly did not fit within the framework established then by *Wallach*[16,17] for the constitution of the monoterpenes. One would have thought that this situation might have led *Ruzicka* toward active involvement with the isoprene rule. However, the following circumstances show how obscured from view were the strands that, woven together, should have led him toward that involvement.

In *Ruzicka*'s 1917 paper on fenchone[19] we find no hint as to the special significance of the structure of that compound. That came much later: in a publication dated 1935 (i.e. ten years after his seminal paper on the isoprene rule) in which, as proof of structure (together with *Reichstein*) of artemisia ketone he described the synthesis of its tetrahydro derivative and stated[25]:

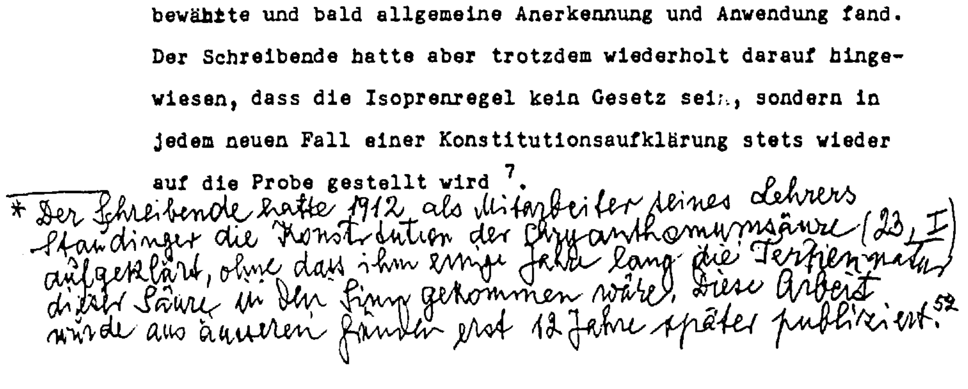
> *«Fenchone and artemisia ketone*[26] *are the only two monoterpene compounds in which the construction of the molecular framework by irregular linkage of isoprene units has been established with certainty.»*

As for the obvious conclusion that this should equally apply to chrysanthemic acid (see Fig. 2) whose structure he, together with *Staudinger*, had solved 20 years earlier, we find no inkling in that paper[4*]; and that in face of the fact that it was the work on that compound (via the derived caronic acid) which must have first made *Ruzicka* aware of the monoterpene literature. In other words, as *Ruzicka* ruefully admitted much later (1963)[24]:

> *«The undersigned had, in 1912, under the guidance of his mentor Staudinger, solved the problem of chrysanthemic acid and for years thereafter failed to notice the terpenic nature of this compound»* (see Fig. 3 and Fig. 4).

Any analysis of *Ruzicka*'s train of thought on the isoprene rule must necessarily take into account the work of *Otto Wallach*. *Wallach*'s 1910 Nobel Prize lecture[16] on his life's work on alicyclic compounds summarized his views on monoterpene constitution as follows:

> *«All recent experiments had already confirmed the accuracy of an older assumption, namely that the hydrocarbons $C_{10}H_{16}$ occurring naturally, are related to a benzene*

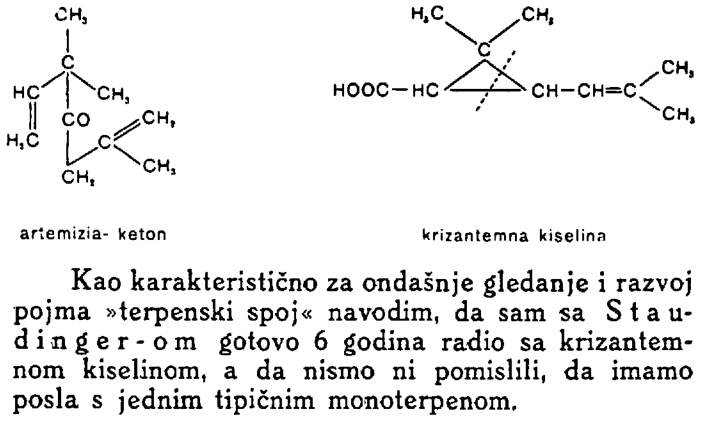


bewährte und bald allgemeine Anerkennung und Anwendung fand. Der Schreibende hatte aber trotzdem wiederholt darauf hingewiesen, dass die Isoprenregel kein Gesetz sei, sondern in jedem neuen Fall einer Konstitutionsaufklärung stets wieder auf die Probe gestellt wird [7].

* Der Schreibende hatte 1912 als Mitarbeiter seines Lehrers Staudinger die Konstitution der Chrysanthemumsäure (23, I) aufgeklärt, ohne dass ihm einige Jahre lang die Terpennatur dieser Säure in den Sinn gekommen wäre. Diese Arbeit wurde aus äusseren Gründen erst 12 Jahre später publiziert.[52]

Fig. 3. From the manuscript for [24].

[4*] In «The Isoprene Rule and the Biogenesis of Terpenic Compounds»[5], published 18 years later, there is likewise no mention of chrysanthemic acid, whereas fenchone and artemisia ketone are mentioned. In view of *Ruzicka*'s difficult personal relationship with his mentor *Staudinger* (cf.[1]) one is tempted to assume that this is a case of Freudian repression. It took *Ruzicka* until 1940 to mention chrysanthemic acid in connection with the isoprene rule – in the Zagreb version of his Nobel lecture (see Fig. 4) held there because of the outbreak of the war; to be omitted again in the 1945 Zürich version of the same lecture[46]. Thereafter it is given full justice in his 1959 Faraday lecture[27], two years after his retirement.



Kao karakteristično za ondašnje gledanje i razvoj pojma »terpenski spoj« navodim, da sam sa Staudinger-om gotovo 6 godina radio sa krizantemnom kiselinom, a da nismo ni pomislili, da imamo posla s jednim tipičnim monoterpenom.

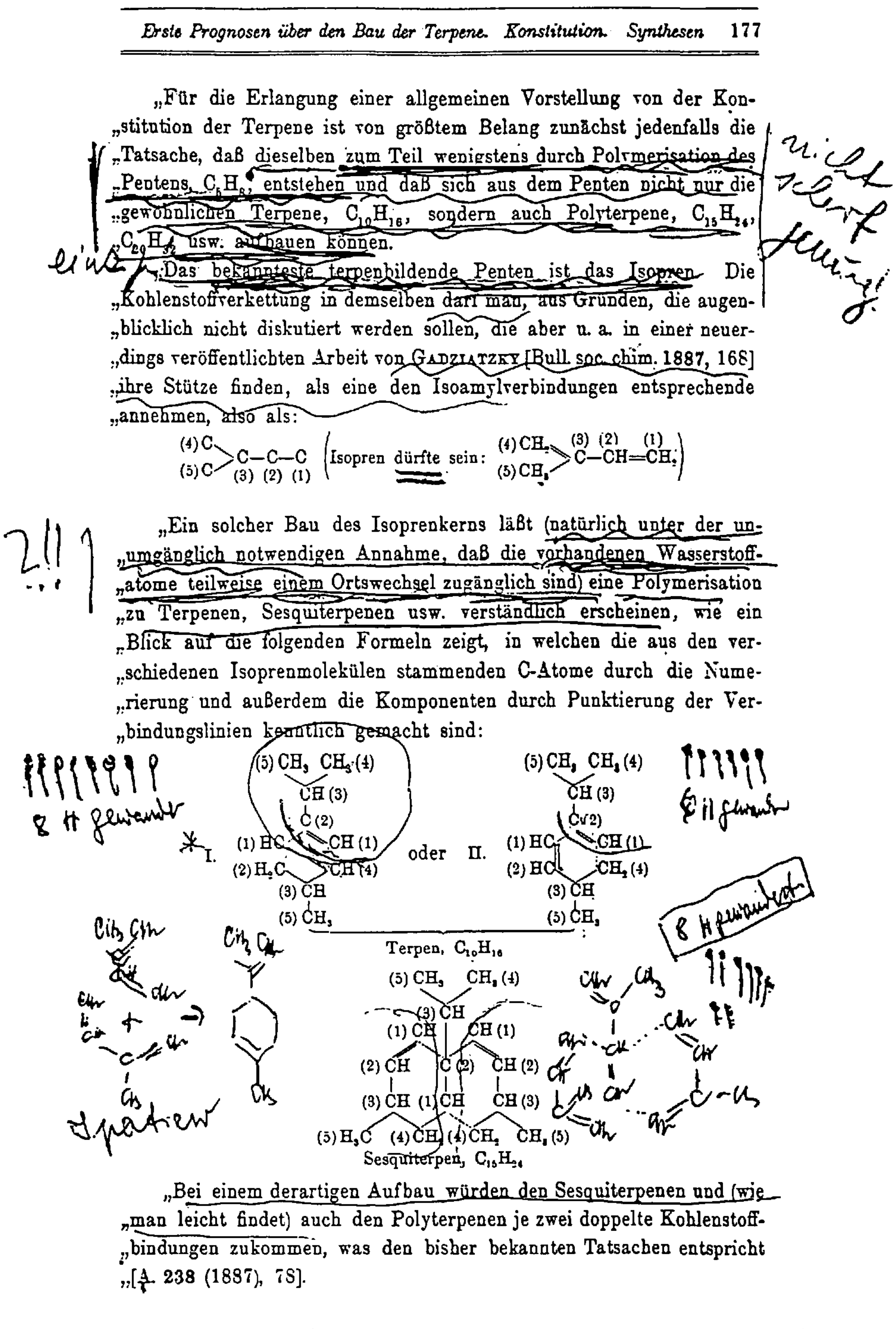
Fig. 4. Reproduction from Lavoslav Ruzicka, «Od dalmatinskog buhaca do seksualnih hormona», Lijecnicki Vjesnik (Zagreb) 62 (1940) 1 (Nobel lecture given in Croatian in Zagreb on 14 March 1940, cf. footnote [7*]*).*

hydrocarbon $C_{10}H_{14}$, the so-called cymol (isopropyl-p-methylbenzene). Pinene, camphor, and other compounds can be converted by appropriate chemical processes to cymol or cymol derivatives, e.g. carvacrol.»

Not a word here of either isoprene or of the isoprene rule. Twenty years earlier *Wallach* had expressed some bold views on the constitutional link between isoprene and the «sesquiterpenes and polyterpenes»[28], but now it seemed that *Wallach*, the cautious experimentalist, did not consider them significant enough for inclusion in his Nobel Prize lecture. On the other hand, one year previous to this in his book «Terpenes and Camphor»[17] he went to great lengths to reiterate his previous views on the subject. Fig. 5 demonstrates this by reproduction from *Ruzicka*'s own personal copy (with the latter's hand-written comments). These two pages not only show the essence of *Wallach*'s ideas on constitutional relationships among the terpenes, they also vividly document *Ruzicka*'s appreciation of his great predecessor whose life and work were the subject of his 1932 Chemical Society Pedler lecture[29]. There is little doubt that in his particular personal attributes, those of courage and imagination in putting forward daring working hypotheses while adamantly insisting on solid experimental proof, *Ruzicka* saw in *Wallach* a kindred spirit.

In his search for a synthesis of irone (see Fig. 2 and Note[2*]) *Ruzicka* had performed the reaction of fenchone with methylmagnesium iodide[12] and, after dehydration of the product, obtained «a hydrocarbon» which, upon ozonolysis, yielded not only fenchone, as might have been expected, but also camphor. This important observation started off his studies of the Wagner rearrangement (first so named by him)[30] and of the structure of «pinene». These studies were indeed relevant to what were at that time urgent problems but there are no indications of any kind that they brought him nearer to the isoprene rule. They did serve as basis for his «Habilitation» dissertation (submitted in 1918), and for the second of his classical achievements, in 1919, the total synthesis of linalool (with *V. Fornasir*, his first doctoral student)[31]. It is also likely that they contributed two years later to the beginning of his lifelong and mutually valued association with the Chuit & Naef Co. (later Firmenich & Co.) in Genève[1,7].

In one of his retrospective lectures[24] *Ruzicka* divided the history of the isoprene rule into three periods, the first of which, spanning the time between *Wallach* and the 1920's, laid the groundwork for the systematic study of the monoterpenes.



Erste Prognosen über den Bau der Terpene. Konstitution. Synthesen 177

„Für die Erlangung einer allgemeinen Vorstellung von der Konstitution der Terpene ist von größtem Belang zunächst jedenfalls die „Tatsache, daß dieselben zum Teil wenigstens durch Polymerisation des „Pentens, C_5H_8, entstehen und daß sich aus dem Penten nicht nur die „gewöhnlichen Terpene, $C_{10}H_{16}$, sondern auch Polyterpene, $C_{15}H_{24}$, „$C_{20}H_{32}$ usw. aufbauen können.

„Das bekannteste terpenbildende Penten ist das Isopren. Die „Kohlenstoffverkettung in demselben darf man, aus Gründen, die augen„blicklich nicht diskutiert werden sollen, die aber u. a. in einer neuer„dings veröffentlichten Arbeit von Gadziatzky [Bull. soc. chim. 1887, 168] „ihre Stütze finden, als eine den Isoamylverbindungen entsprechende „annehmen, also als:

$$\left.\begin{matrix}(4)\,C\\(5)\,C\end{matrix}\right>\underset{(3)}{C}-\underset{(2)}{C}-\underset{(1)}{C}\quad\left(\text{Isopren dürfte sein: }\left.\begin{matrix}(4)\,CH_3\\(5)\,CH_3\end{matrix}\right>\overset{(3)}{C}-\overset{(2)}{CH}=\overset{(1)}{CH_2}\right)$$

„Ein solcher Bau des Isoprenkerns läßt (natürlich unter der unumgänglich notwendigen Annahme, daß die vorhandenen Wasserstoff„atome teilweise einem Ortswechsel zugänglich sind) eine Polymerisation „zu Terpenen, Sesquiterpenen usw. verständlich erscheinen, wie ein „Blick auf die folgenden Formeln zeigt, in welchen die aus den ver„schiedenen Isoprenmolekülen stammenden C-Atome durch die Nume„rierung und außerdem die Komponenten durch Punktierung der Ver„bindungslinien kenntlich gemacht sind:

„Bei einem derartigen Aufbau würden den Sesquiterpenen und (wie „man leicht findet) auch den Polyterpenen je zwei doppelte Kohlenstoff„bindungen zukommen, was den bisher bekannten Tatsachen entspricht „[A. 238 (1887), 78].

Fig. 5. From Ruzicka's personal copy of Otto Wallach's «Terpene und Campher»[17].

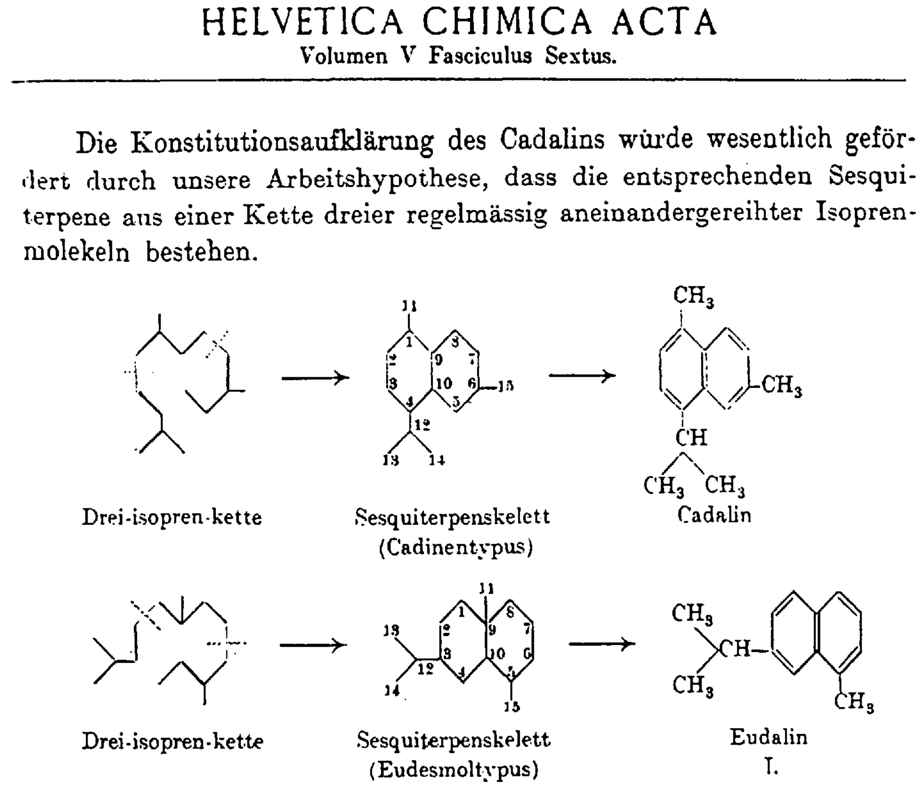
Concerning the sesquiterpenes he wrote as follows:

«In 1887 Wallach had suggested that monoterpenes were formed by combination of 2 isoprene units, and had tentatively extended this to proposing a basis of three such units for the sesquiterpenes. The first two sesquiterpenes whose structures were then established unequivocally: α-santalene (Semmler, 1910) and farnesol (Kerschbaum, 1913) are indeed constructed from three isoprene units, but that fact seemed to have escaped notice altogether. At any rate there was certainly no such thing then as an isoprene rule serving as a working hypothesis for structure elucidation.»

Any objective examination of the terpene literature of that period must conclude that these comments depicted a true picture of the situation (see also Fig. 5). Moreover, there is no doubt that it was from then on (1920)[32] that the work of *Ruzicka* and his co-workers, based on the use of dehydrogenative degradation in structure determination of sesquiterpenes of the cadalene and eudalene series, led to the breakthrough toward what later was termed the empirical isoprene rule (cf. Fig. 6). The crucial statement, dating from 1922[33] reads as follows:

«The main conclusion so far from our sesquiterpene work is that practically all compounds whose carbon skeleton is now known show a close mutual relationship, being derivable from a regular three-isoprene chain, as produced in nature in the case of farnesol.»

It is striking how the analogy in molecular build between the cyclic mono- and the sesquiterpenes is reflected in the way their constitution was uncovered. In either case recourse to studying their relationships with the familiar corresponding aromatic systems was essential. In one case it was *p*-cymene, in the other cadalene and eudalene. It was fortunate, and is in retrospect quite significant, that it was those terpenes that had a *regular* isoprene arrangement which at the beginning served to reveal such a relationship[5*].

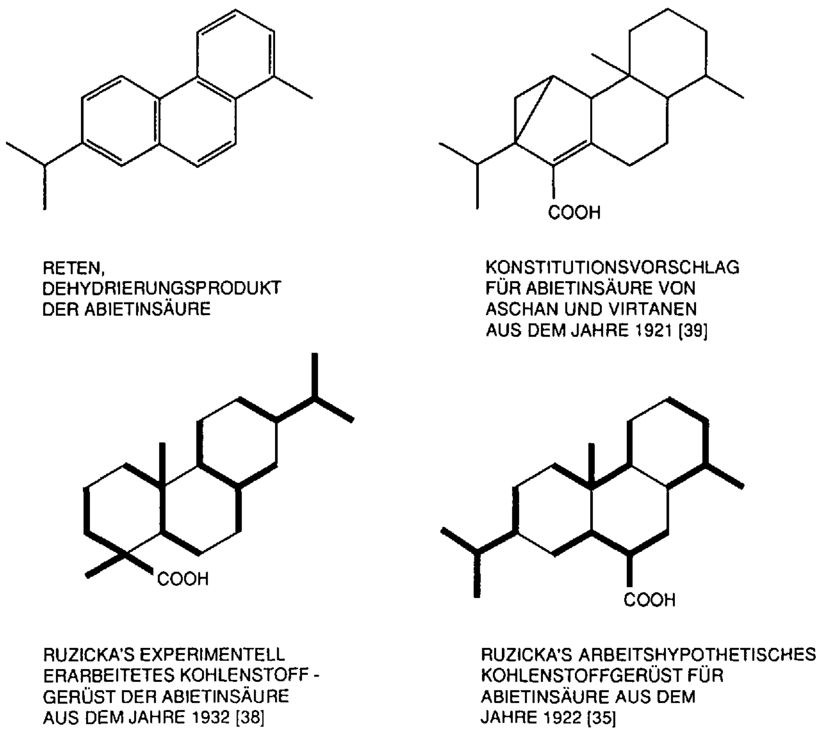


HELVETICA CHIMICA ACTA
Volumen V Fasciculus Sextus.

Die Konstitutionsaufklärung des Cadalins würde wesentlich gefördert durch unsere Arbeitshypothese, dass die entsprechenden Sesquiterpene aus einer Kette dreier regelmässig aneinandergereihter Isoprenmolekeln bestehen.

Fig. 6. From Ruzicka's seminal papers on the isoprene rule, 1922 [33].

It was characteristic of *Ruzicka* that he did not leave his conclusions to rest in the form of a «Cadalene and Eudalene Rule» applicable merely to that particular corner of sesquiterpene chemistry, but that he advanced from there toward the isoprene rule to serve as a generally valid aid to structure determination of the higher terpenes. Of decisive importance in this regard, as he himself pointed out[27], was his concurrent work on abietic acid[35], a diterpene acid the structure of whose dehydrogenation product retene (see Fig. 7) was then already known. The deductive path leading from the latter to that of the natural product itself was here much more difficult to traverse and took more than ten years[38] – no wonder, since abietic acid is a diterpene



RETEN, DEHYDRIERUNGSPRODUKT DER ABIETINSÄURE

KONSTITUTIONSVORSCHLAG FÜR ABIETINSÄURE VON ASCHAN UND VIRTANEN AUS DEM JAHRE 1921 [39]

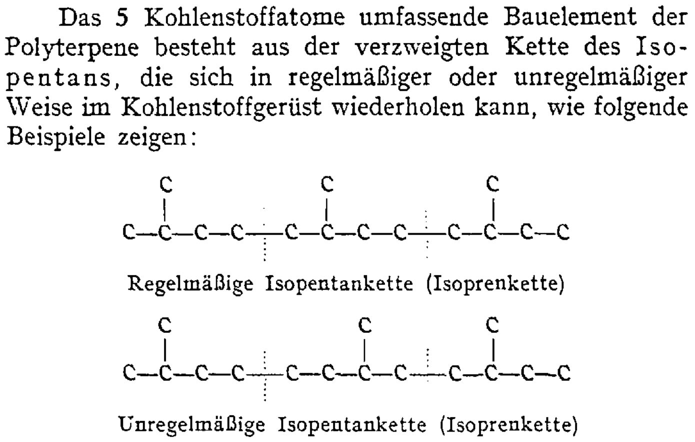
RUZICKA'S EXPERIMENTELL ERARBEITETES KOHLENSTOFF-GERÜST DER ABIETINSÄURE AUS DEM JAHRE 1932 [38]

RUZICKA'S ARBEITSHYPOTHETISCHES KOHLENSTOFFGERÜST FÜR ABIETINSÄURE AUS DEM JAHRE 1922 [35]

Fig. 7. On the role of the isoprene rule in the determination of constitution of abietic acid.

[5*] The term «regular isoprene chain» («head-to-tail union of isoprene units» according to *Robert Robinson* [34]) was meant to describe that isoprene unit arrangement to be found in the *aliphatic* mono- and sesquiterpenes (Fig. 8) as well as in natural rubber (see *Harries* [36], 1914). In the case of natural rubber this arrangement was interpreted as resulting from linking of isoprene molecules at their 1- and 4-positions, in line with *Thiele*'s theory of partial valencies. Incidentally, it seems of historical significance that *Ruzicka*'s seminal papers on the constitution of the sesquiterpenes[33], and *Staudinger*'s important publication on the structure of natural rubber[37] appeared in the same 1922 volume of Helvetica Chimica Acta. To be sure, *Ruzicka* did not create the isoprene rule as such; what he did was to recognize and to demonstrate its importance for the structure determination of the cyclic sesqui-, di-, and triterpenes.

with an *irregular* isoprene pattern. Fig. 7, showing *Ruzicka's* initial working hypothesis as compared with the finally established structure, hints at how difficult it was to arrive at the correct solution once it became clear that the isoprene chain arrangement was not a regular one (Fig. 8).



Das 5 Kohlenstoffatome umfassende Bauelement der Polyterpene besteht aus der verzweigten Kette des Isopentans, die sich in regelmäßiger oder unregelmäßiger Weise im Kohlenstoffgerüst wiederholen kann, wie folgende Beispiele zeigen:

Regelmäßige Isopentankette (Isoprenkette)

Unregelmäßige Isopentankette (Isoprenkette)

Fig. 8. From the introduction to Ruzicka's review article «Die Architektur der Polyterpene» [45].

The Isoprene Rule as an Instrument for Structure Elucidation

Another aspect of Fig. 7 is the way it illustrates the manner in which *Ruzicka* initially utilized the isoprene rule – this from the juxtaposition of his own 1922 ideas with the 1921 structure for abietic acid suggested by *Aschan* and *Virtanen* [39] which, as specially emphasized in his Faraday lecture[27], was at variance with the rule. For the whole of what he termed the rule's «second period», from 1921 to 1953, it was basically a device for the formalistic dissection of a carbon skeleton into isopentane units. No thought was given to what implications this might have on the question of how in actual fact these molecules were constructed by Nature.

For us in this day and age, having been conditioned by half a century of mechanistic enlightenment and biosynthetic insight, we find it difficult to look at structures of natural products from the perspective of a classical terpene chemist. We tend to forget that he in turn had been conditioned by the fact that essentially nothing was known

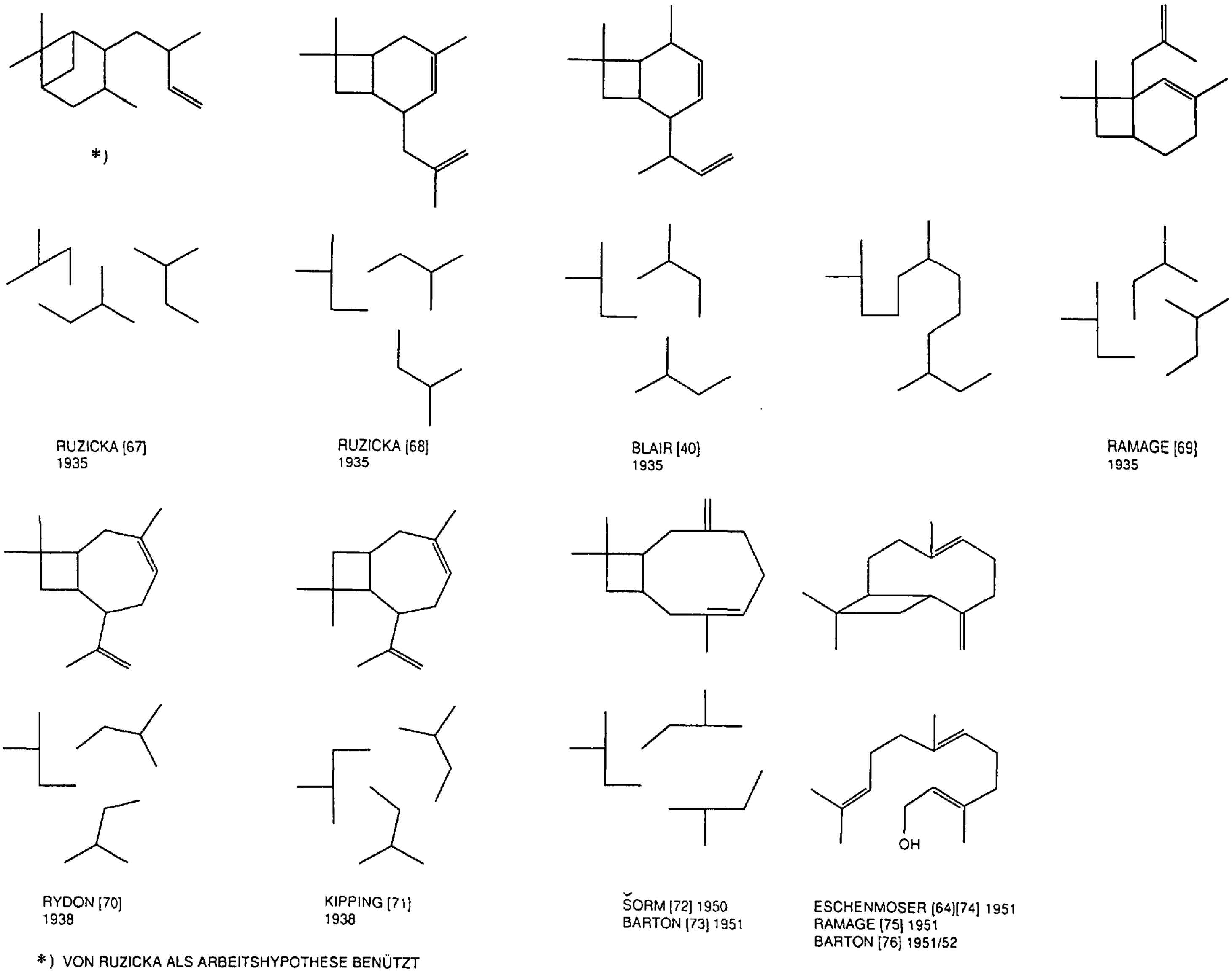


Fig. 9. The various constitutional formulae put forward in connection with the elucidation of constitution of β-caryophyllene.

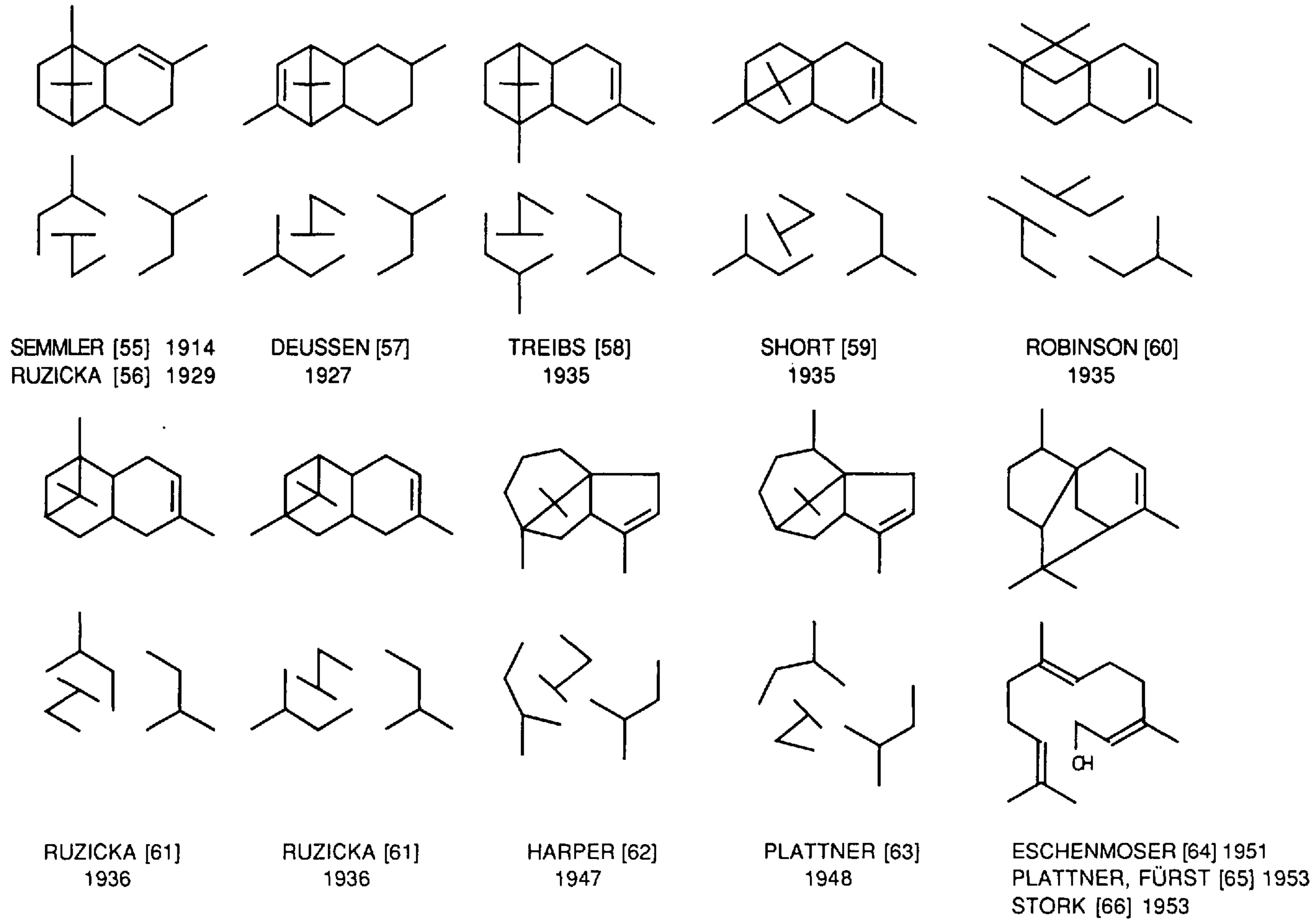


Fig. 10. The various constitutional formulae put forward in connection with the elucidation of constitution of cedrene.

about biosynthesis of natural products, and by the view that natural processes in biosynthesis might quite conceivably proceed by reaction types fundamentally different from those known to chemists. The more we realize this fact, the better can we comprehend *Ruzicka*'s elation when the curtain began to rise on the «third period», that of the biogenetic isoprene rule, as brought about by better understanding of reaction mechanisms in organic chemistry and the advent of lanosterol.

Well into the 1950's there is no lack of examples in the literature, both by *Ruzicka* and by other workers in the field, of the purely formalistic use of the isoprene rule. Here and there, however, we do find attempts to break out of this mold, particularly where sesquiterpenes such as β-caryophyllene and cedrene were concerned, and where dehydrogenative degradation was of little use. Figs. 9 and 10 show the variety of often weird working hypotheses suggested by various groups during that period for these two natural products. All of them took the isoprene rule into account but were in retrospect little more than topological manipulations without regard to mechanistic reasoning. This was true not just for *Ruzicka* but even for someone like *Robert Robinson*, the champion of his time in the field of organic reaction mechanisms.

None of the early hypotheses suggested that both these sesquiterpenes exhibit a regular (i.e. head-to-tail) three-isopentane array, as had been demonstrated in 1922 for the classical sesquiterpenes of the cadalene and eudalene type (see Fig. 6). One exception was that put forward in 1935 by *Blair*[40] who used this very criterion in the case of caryophyllene, on the basis of a comment by *Ingold*[41], but still for purely topological reasons. *Ruzicka*'s reaction[42] to this is illustrative of what in 1936 were his views:

> *«Blair has written 'it was pointed out by Ingold (Ann. Reports Chem. Soc., 1924, 103) that the sesquiterpene skeleton is not merely composed of three isoprene units but is, so to speak, a folded farnesol chain'. He has overlooked the fact that our early work on the carbon skeleton of the sesquiterpenes of cadalene and eudalene type had derived these from farnesol; cf., Helv. Chim. Acta, 1922, 5 534, 968, where these conclusions were clearly expressed by structural formulae»* ([42], *footnote on p. 348).*

With this remark, relegated to footnote status, *Ruzicka* claims priority for the idea of regular isoprene connectivity while at the same time (but now in the text of the paper) he is explicit about the limitations:

> *«It is true that at the time of our work on cadalene and eudalene type sesquiterpenes we had derived these structures from farnesol, and that subsequent to this we were able to suggest further examples confirming this scheme of construction. However, we do not think it permissible to apply it a priori to all sesquiterpenes. In all other sub-groups there are cases where the rule of regular isoprene unit construction is not valid, such as in the cases of fenchone and camphor among the monoterpenes, abietic and dextropimaric acids among the diterpenes, and the pentacyclic triterpenes. One must on no account cling to this rule as a working hypothesis where it is not supported by clear and unambiguous degradation results.»*

This little altercation is significant not only because it again manifests *Ruzicka*'s thinking at the time but also because we see in *Blair*'s proposal an inkling of the biogenetic isoprene rule to appear fifteen years later. By then both caryophyllene and cedrene would indeed be seen as derived from a regular three-isoprene arrangement as in farnesol, but mainly this was to come about through a new conception of the rule, not as a result of new degradation data, and above all from an insight into reaction mechanisms, for which the time was not yet ripe in the 1930's. *Ruzicka* had in fact taken the first step in that direction already in 1925, in connection with his work on the acid-catalyzed cyclization of the sesquiterpene nerolidol to give products of the monocyclic bisabolene type. At the time he commented that «perhaps such a transformation of acyclic precursors into cyclic sesquiterpenes could similarly occur in nature»[43][6*]. However, as we see from his 1935 retort to *Blair*, he had become a great deal more cautious in the interim. What in retrospect he was to call the «Farnesol Rule»[5] was not yet to be developed into the refined tool of structure determination which it was destined to become. The problems he had with caryophyllene, cedrene, and elemol hindered, rather than helped, in this respect. Moreover, the more complex di- and triterpenes, which by then he was working on at full steam, were convincing him to an increasing degree of the overriding importance of experimental work, since he was being confronted with numerous carbon skeletons clearly based on *irregular* isoprene arrangements.

These circumstances may account to some extent for the relative lack of progress in the development of the rule during the period 1925–1935, but they were not the only reason. *Ruzicka*'s interests were then steadily veering toward subjects, unconnected with the isoprene rule, that propelled him into public attention and esteem. These were the large-ring compounds and odoriferous principles of the musk type (from 1924) and steroids and male sex hormones (from 1933). In the meantime he was basking in the satisfaction of seeing how his Zürich colleague and «rival» *Paul Karrer* had to accord serious attention to the isoprene rule in his work on the biologically important squalene, vitamin A and the carotenoids[45].

By 1945 *Ruzicka* had attained a matured and somewhat detached attitude toward the isoprene rule, best expressed in his Nobel Prize lecture[46][7*]:

«Even if on occasion we were to encounter a natural product whose structure does not strictly obey the isoprene rule, that would not at all diminish its importance as a working hypothesis. Experience has taught us that there are no rules of molecular construction to which there are no exceptions, and that we are duty bound to examine the applicability of such rules in each and every case irrespective of whether their validity had in the past seemed absolute or whether exceptions had already been found.»

«It is worth considering whether the isoprene rule is valid not only as a working hypothesis in the laboratory, but beyond that also as a basis of the workings of Nature. The regularities observed in the structures of the higher terpenes point to the likelihood of similarly regular and uniform principles of construction by Nature, albeit in the absence so far of any clue as to the mechanism of such a widespread biochemical pathway.»

Time for Change

In retrospect the above observations seem prophetic. They date from a period during which the first steps had already been taken in *Ruzicka*'s laboratory toward the structure elucidation of lanosterol[47]. The two guidelines expressed above: the basically *conditional* validity of the empirical isoprene rule, and the absolute priority to be accorded to experimental results, were to prove their worth later, on the occasion of the victory of the Ruzicka-Jeger group[48] in their race with *Barton*[49] to finally solve the lanosterol structure. It emerged that it was the carbon skeleton of that particular natural product, of fundamental biological importance, which could *not* be perceived in terms of isoprene unit construction at all[8*]. And as for *Ruzicka*'s expectations that the isoprene rule would emerge as a harbinger of our conception of the way Nature constructs terpene molecules, it was at that very time that the «count-down» had begun: *Bloch* and *Rittenberg* found in 1945[52], using ^{14}C-isotope tracers, that cholesterol is biosynthesized from acetic acid.

In their search for constitutional relationships, to serve in understanding the way molecules are constructed by Nature, there is much similarity between *Ruzicka* and his great British contemporary *Robert Robinson*. What *Ruzicka* did from 1920 onwards in the terpene field was paralleled by *Robinson*'s work starting in 1917[53] in the field of alkaloids which occupied the central position in natural product chemistry at that time. *Robinson* summarized his decades of achievement in 1953 in the notable Weizmann Memorial Lectures[53b]; in that same year *Ruzicka* delivered his lecture on the biogenetic isoprene rule at the IUPAC congress in Stockholm[5]. *Robinson* had the upper hand in his intuitive insight on reactivity and mechanism[54] and in perceiving biosynthetic relationships between natural products, especially alkaloids. But, in a comparison of the relevance of their work to the chemistry of life, it was *Ruzicka*, the experimentalist, working on the terpenes which in all their variety turned out to be of biologically greater importance than the alkaloids, who was in the end to have the best of it. Whatever he may have lacked in mechanistic insight he made up for by the fact that by the time he reached the critical early 1950's he was not left to fend for himself, but had managed to establish a framework of cooperation with the younger generation in his circle.

At the end of the second world war there was the period during which *Ruzicka* had began to detach himself from chemistry to an increasing degree, in order to devote his efforts to organizations established to alleviate postwar hardship, particular in his native Yugoslavia. In addition, he began to invest the considerable sums accumulated during the war from industrial utilization of his research in his notable collection of Dutch paintings. All of this he did with the same singleness of purpose that had been characteristic of his scientific endeavours. As a result he was often away from the laboratory and out of touch with his doctoral students. Nonetheless, his institute continued to function perfectly, thanks to the capable hands of *Vlado Prelog* and *Placidus Plattner* who had become associate professors in 1949. The triterpene work which had been dearest to his heart was continuing at full steam under the increasingly independent direction of *Oskar Jeger*. *Andor Fürst*, together with *Plattner* who was looking after the administrative side, were continuing with sesquiterpenes in which *Ruzicka*'s interest had waned considerably, while both *Hans Heusser* and *Plattner* were busy with steroids. *Hans Günthard* had been given the task of establishing infra-red spectroscopy in the institute, and *Emil Hardegger* had branched out into the sugars. As for the monoterpenes, in particular odoriferous substances, *Ruzicka* – having at long last solved the irone problem – left this field in the hands of his veteran associates *Hans Schinz* and *Casimir Seidel*.

It was to *Schinz*'s special credit that he used the freedom *Ruzicka* had granted him[9*] to embark on a systematic study of the acid-catalyzed cyclization of polyenes using model systems related to the mono- and sesquiterpenes, a reaction type discovered by *Tiemann*[77]. It was this work which first brought the present author into contact with this reaction in his own diploma and doctoral work, leading to years of study on the mechanism and steric course of polyene cyclization and therefore, in a way, contributing to the decision to write this article in the first place.

The year 1951 marked *Ruzicka*'s chemical «renaissance». It was also then that he decided to re-marry. The concurrent return to chemistry came just in time. Together with *Prelog* and *Plattner*[10*], both fresh from their first visit to the USA, a new generation with a new outlook on

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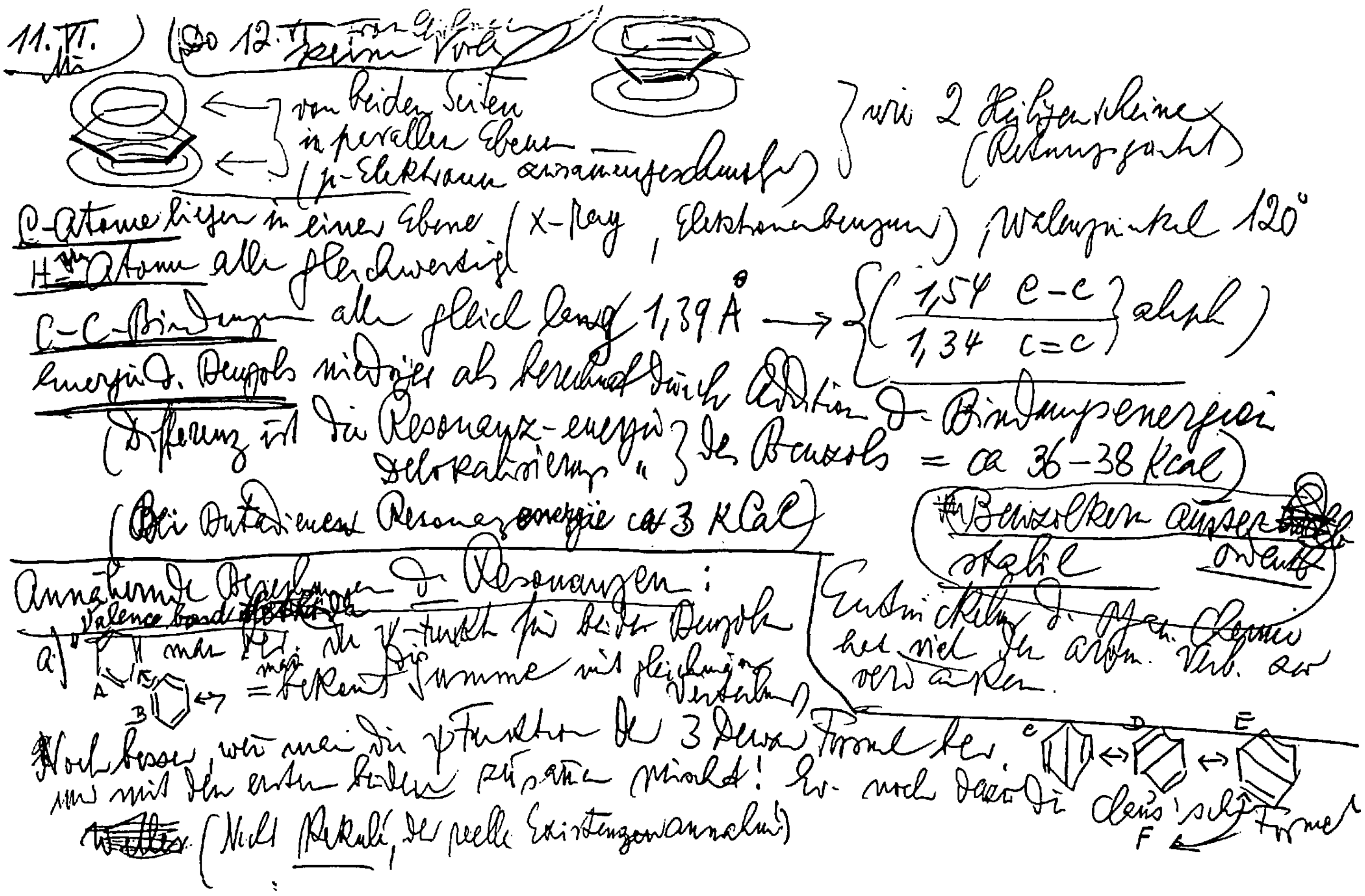
[6*] For a later, and in retrospect more extended proposal cf.[44].

[7*] *Ruzicka* was awarded the Nobel Prize ("for his work on polymethylenes and higher terpenes") in 1939, but because of the war his (official) Prize lecture had to be postponed until 1945 (in Zürich).

[8*] This was the second example of a terpene whose constitution contradicted the empirical isoprene rule. The first was eremophilone whose constitution was solved by *Simonsen* in 1937[50]. Here *Robinson*[51] suggested that its carbon skeleton could be derived from a normal one by assuming a 1 → 2 methyl shift. This was later considered by *Ruzicka* to have been a first step toward the biogenetic isoprene rule.

[9*] *Schinz* was an external employee of Firmenich & Co. in Genève. He took his doctorate with *Ruzicka* in 1924, remained in his group at the ETH, and toward the end of the war was permitted to work independently with doctoral students.

[10*] In 1952 *Plattner* left the Organic Chemical Laboratory and assumed the position of Director of Research in Basel.



11.VI. (bis 12.VI)

von beiden Seiten in parallelen Ebenen (p-Elektronen zusammengedrückt) } wie 2 [illegible] (Rettungsgürtel)

C-Atome liegen in einer Ebene (X-Ray, Elektronenbeugung) Valenzwinkel 120°

H-Atome alle gleichwertig

C-C-Bindungen alle gleich lang 1,39 Å → {(1,54 C–C / 1,34 C=C) aliph.)

Energieinh. Benzols niedriger als berechnet durch Addition d. Bindungsenergien

(Differenz ist die Resonanz-energie „Delokalisierungs" } des Benzols = ca 36–38 Kcal)

(Bei Butadienen Resonanzenergie ca 3 KCal)

(Benzolkern ausserordentl. stabil)

Annähernde Berechnungen d. Resonanzen: ~~Valence bond Methode~~

a) [illegible] die ψ-Funkt. für beide Benzole = bekannt Summe mit gleichmässiger Verteilung

A B

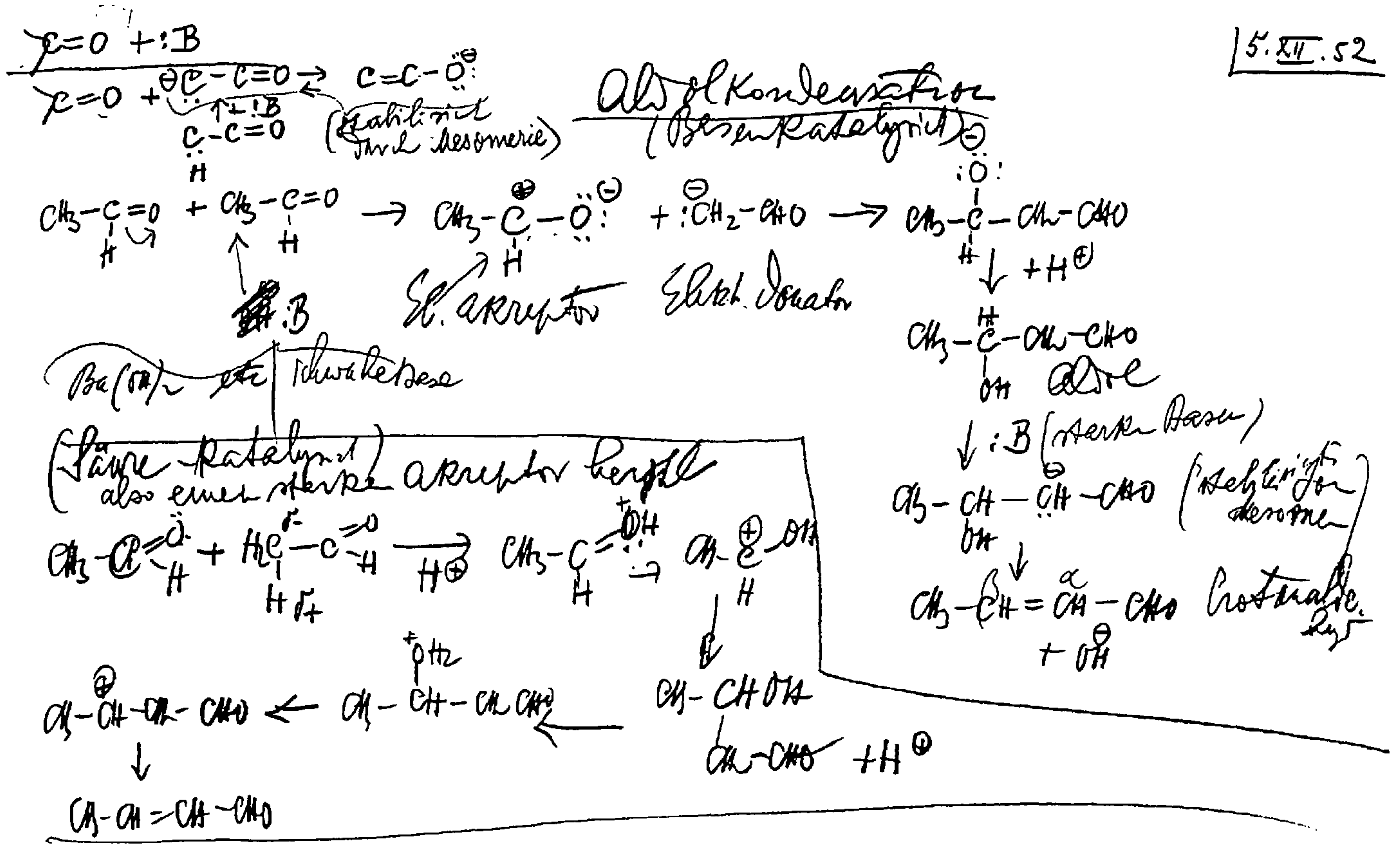
Entwicklung d. org. Chemie hat viel zu arom. Verb. zu verdanken.

Noch besser, wenn man die ψ Funktion der 3 Dewar Formel bei, nur mit den ersten beiden zusammen mischt! Evtl. noch dazu die Claus'sche Formel

C D E F

(Nicht Kekulé, der reelle Existenzannahme)

Fig. 11. Excerpt from Ruzicka's notes on Prelog's lecture «Organic Chemistry I», summer term 1952.



5.XII.52

Aldolkondensation (Basenkatalysiert)

C=O + :B

C=O + ⊖C–C=O ⇌ C=C–O⊖ (stabilisiert durch Resonanz)

El. akzeptor

Elektr. Donator

Ba(OH)2 etc schwache Base

Aldol

:B (starke Base)

Crotonaldehyd

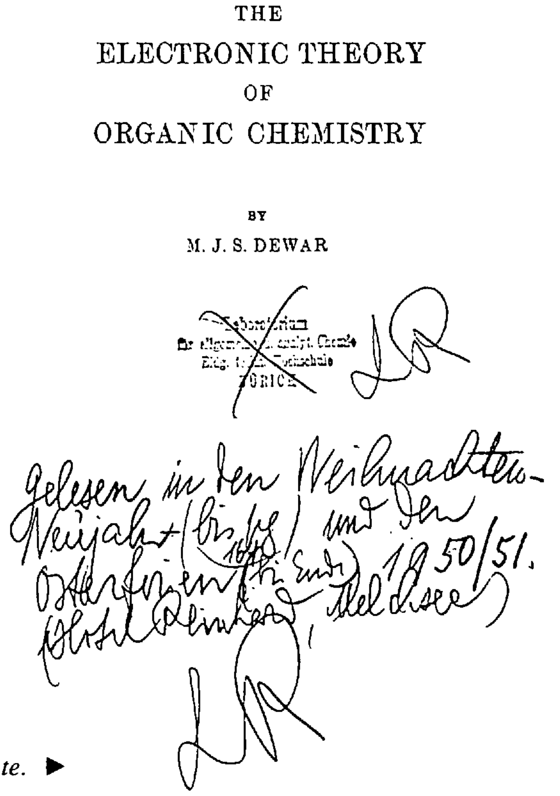
~~Säure Katalysiert~~ also einen starken akzeptor benötigt

Fig. 12. Excerpt from Ruzicka's notes on Prelog's lecture «Organic Chemistry II», winter term 1952/53.

organic chemistry was entrenching itself at the ETH; the age of reaction mechanism and of the electronic theory of organic chemistry had begun. The work on the constitution of lanosterol was reaching its climax. In the field of the pentacyclic triterpenes of by then known structure, *Jeger* had in the preceding year put forward a first, albeit hesitant, hypothesis on a possible biogenetic connection between the carbon skeletons of β- and α-amyrin, lupeol, and germanicol[78]. In the sesquiterpene field the present author had used a theory of the mechanism of acid-catalyzed polyene cyclization to derive new structures for cedrene, β- and α-caryophyllene, lanceol, and elemol (see Fig. 9 and Fig. 10) from the structure of farnesol or the related farnesene, and proposed that such mechanistic derivation of the constitution of terpenes represents an additional selection criterion for using the isoprene rule in structure determination of such compounds[64]. Finally, during that year *Bloch* in Chicago was on his way to the proof, published a year later, that squalene is an intermediate in the biosynthetic conversion of acetic acid to cholesterol[79].

It was remarkable how *Ruzicka,* then aged 64, reacted to these developments. During the summer semester of 1952 and the winter semester of 1952/53 he was to be found on most days sitting and listening attentively to *Prelog*'s course – organic chemistry I and II – for beginners. It goes without saying that this was not, God forbid, to keep a watch on *Prelog*'s teaching, but in order to take in a new outlook on organic chemistry. Among the papers left by *Ruzicka* we can find a document of a kind singular in the history of the Institute of Organic Chemistry at the ETH Zürich (and probably in that of any other university): *Prelog*'s basic course in Organic Chemistry, painstakingly taken down in *Ruzicka*'s handwriting on carefully numbered and dated loose sheets of notepaper, as exemplified by Figs. 11 and 12.

The new *Ruzicka* was not slow in drawing his conclusions from the impact of the new age of reaction mechanism on the fate of the isoprene rule (perhaps with the impetus of developments in the sesquiterpene field). This is vividly clear from Figs. 13 and 14: during the winter of 1950/51 he made an exhaustive and practically word-for-word study of *Michael Dewar*'s new book[80], then considered sensational (see Fig. 14), to which he devoted his entire Christmas and Easter holidays as we see from Fig. 13.



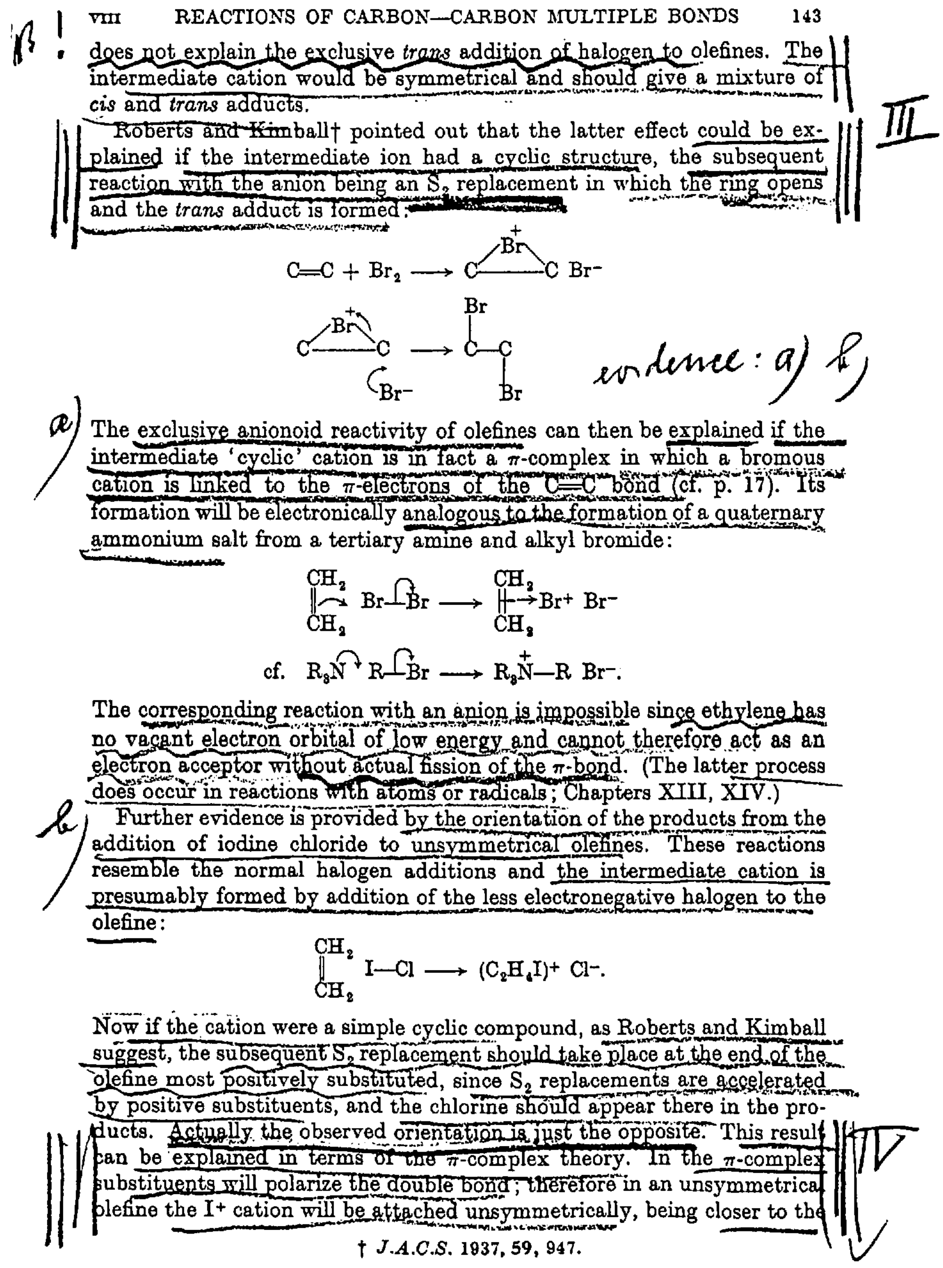
THE
ELECTRONIC THEORY
OF
ORGANIC CHEMISTRY

BY
M. J. S. DEWAR

~~Laboratorium für allgemeine u. analyt. Chemie Eidg. techn. Hochschule ZÜRICH~~

Gelesen in den Weihnachts-Neujahr (bis [illegible]) und den Osterferien (bis Ende) 1950/51. ([illegible], [illegible])

Fig. 13. Title page of Dewar's text book[80]*, 1949, with Ruzicka's note.* ▶



β !

VIII REACTIONS OF CARBON—CARBON MULTIPLE BONDS 143

does not explain the exclusive *trans* addition of halogen to olefines. The intermediate cation would be symmetrical and should give a mixture of *cis* and *trans* adducts.

Roberts and Kimball† pointed out that the latter effect could be explained if the intermediate ion had a cyclic structure, the subsequent reaction with the anion being an S_2 replacement in which the ring opens and the *trans* adduct is formed:

III

$$\mathrm{C{=}C + Br_2 \longrightarrow C\overset{\overset{+}{Br}}{\text{——}}C\ Br^-}$$

$$\mathrm{C\overset{\overset{+}{Br}}{\text{——}}C\ \ Br^- \longrightarrow \underset{}{C(Br)\text{—}C(Br)}}$$

evidence: a) b)

a) The exclusive anionoid reactivity of olefines can then be explained if the intermediate 'cyclic' cation is in fact a π-complex in which a bromous cation is linked to the π-electrons of the C═C bond (cf. p. 17). Its formation will be electronically analogous to the formation of a quaternary ammonium salt from a tertiary amine and alkyl bromide:

$$\mathrm{CH_2{=}CH_2\ \ Br\text{—}Br \longrightarrow CH_2{=}CH_2 \rightarrow Br^+\ Br^-}$$

$$\text{cf.}\quad \mathrm{R_3N\ \ R\text{—}Br \longrightarrow R_3\overset{+}{N}\text{—}R\ Br^-.}$$

The corresponding reaction with an anion is impossible since ethylene has no vacant electron orbital of low energy and cannot therefore act as an electron acceptor without actual fission of the π-bond. (The latter process does occur in reactions with atoms or radicals; Chapters XIII, XIV.)

b) Further evidence is provided by the orientation of the products from the addition of iodine chloride to unsymmetrical olefines. These reactions resemble the normal halogen additions and the intermediate cation is presumably formed by addition of the less electronegative halogen to the olefine:

$$\mathrm{CH_2{=}CH_2\ \ I\text{—}Cl \longrightarrow (C_2H_4I)^+\ Cl^-.}$$

Now if the cation were a simple cyclic compound, as Roberts and Kimball suggest, the subsequent S_2 replacement should take place at the end of the olefine most positively substituted, since S_2 replacements are accelerated by positive substituents, and the chlorine should appear there in the products. Actually the observed orientation is just the opposite. This result can be explained in terms of the π-complex theory. In the π-complex substituents will polarize the double bond; therefore in an unsymmetrical olefine the I^+ cation will be attached unsymmetrically, being closer to the

IV

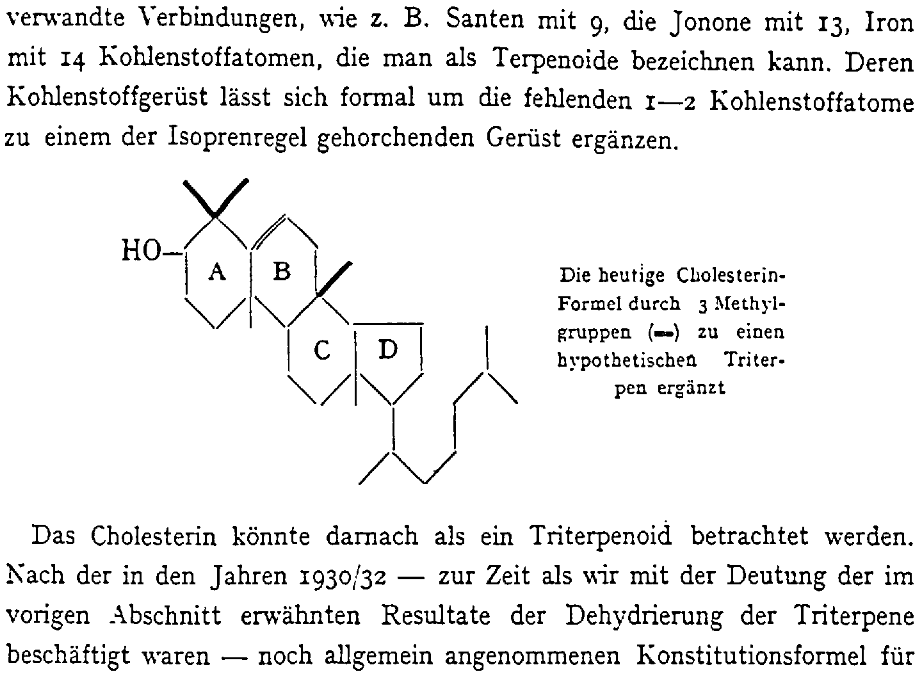
† *J.A.C.S.* 1937, 59, 947.

Fig. 14. A page in Dewar's text book[80] *with characteristic Ruzicka traces.* ▶

«Is Lanostadienol (Lanosterol) a Triterpene or a Steroid?» – this emphatic question was central to the 1952 Jeger-Ruzicka paper[48] presenting proof of structure of this now crucially prominent natural product. Their considered reply was the following:

«In spite of its having 30 carbon atoms, and of the identity of rings A and B with those found in the pentacyclic triterpenes, lanosterol does not obey the isoprene rule and hence cannot be considered a triterpene.»

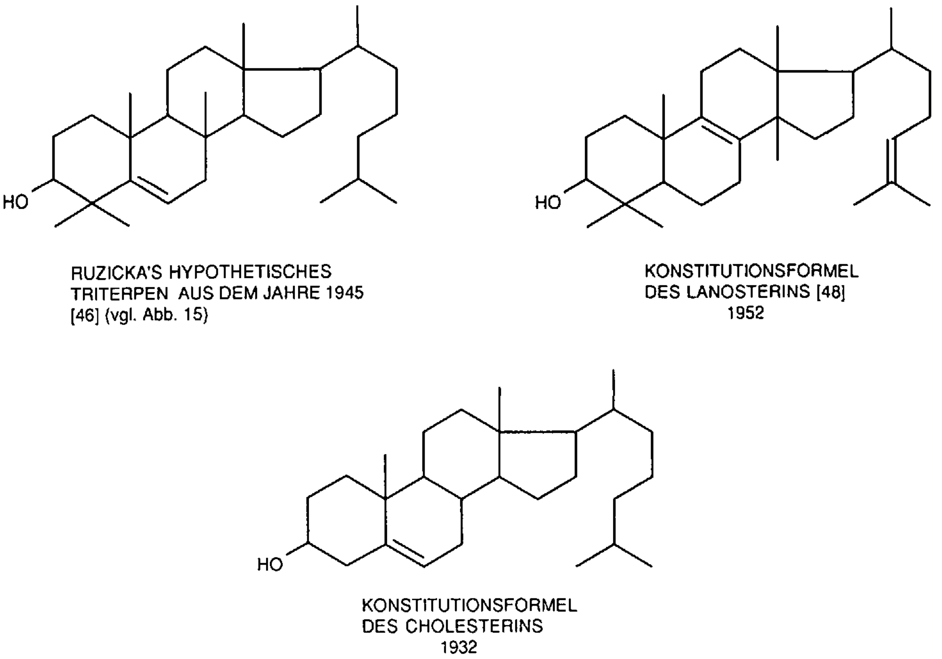
Both question and answer were to represent the last vestiges of a static mental attitude which had typified the «classical» period of triterpene and steroid research. It was to come to an end in the following year when *Woodward* and *Bloch* published their proposal[81], supported by carefully planned experimental work, that lanosterol is an intermediate in the biosynthesis of cholesterol. That began a new era, one in which *Ruzicka* saw himself as having reached a goal which could not have given him grater satisfaction: lanosterol as the link between the two major areas in his life's work: terpenes and steorids. That this possibility had been on his mind for a long time[11*], giving rise to the prescient remarks in his Nobel Prize lecture in 1945[46], is indicated by Figs. 15 and 16.


verwandte Verbindungen, wie z. B. Santen mit 9, die Jonone mit 13, Iron mit 14 Kohlenstoffatomen, die man als Terpenoide bezeichnen kann. Deren Kohlenstoffgerüst lässt sich formal um die fehlenden 1—2 Kohlenstoffatome zu einem der Isoprenregel gehorchenden Gerüst ergänzen.

Die heutige Cholesterin-Formel durch 3 Methylgruppen (▬) zu einen hypothetischen Triterpen ergänzt

Das Cholesterin könnte darnach als ein Triterpenoid betrachtet werden. Nach der in den Jahren 1930/32 — zur Zeit als wir mit der Deutung der im vorigen Abschnitt erwähnten Resultate der Dehydrierung der Triterpene beschäftigt waren — noch allgemein angenommenen Konstitutionsformel für

Fig. 15. Reproduction from Ruzicka's Nobel lecture, 1945[46].



RUZICKA'S HYPOTHETISCHES TRITERPEN AUS DEM JAHRE 1945 [46] (vgl. Abb. 15)

KONSTITUTIONSFORMEL DES LANOSTERINS [48] 1952

KONSTITUTIONSFORMEL DES CHOLESTERINS 1932

Fig. 16. Ruzicka's hypothetical triterpene of 1945 (Fig. 15) compared to constitutional formulae of lanosterol and cholesterol.

The Biogenetic Isoprene Rule

For *Ruzicka* and his group lanosterol was both victory and defeat. The paper by *Woodward* and *Bloch*, interpreting the constitutional relationship between squalene and cholesterol without however providing any mechanistic detail, was to be the first step in a dramatic turn of events in terpene chemistry.

Instinctively *Ruzicka* acted in the right direction. In the first place he tried to induce *Konrad Bloch* to come to the ETH as its first Professor of Biochemistry. In 1953 *Bloch* had spent half a year on sabbatical leave in *Ruzicka*'s institute and had with his own hands provided experimental proof that C-7 in cholesterol was indeed derived from acetic acid, in accord with the squalene conformation *Woodward* had proposed to him[83], and not with *Robinson*'s earlier (1934) suggestion[84]. However, when faced with the choice ETH or Harvard he decided for the latter[85].

The second step was to write and publish the paper on the biogenetic isoprene rule, parallel to the talk he gave that summer at the IUPAC congress in Stockholm[5]. In a chapter of this publication entitled «Biogenesis of Steroids and Terpenic Compounds» he presented, with *A. Eschenmoser* and *H. Heusser* as coauthors, the ideas known at that time on the mechanistic derivation of terpenes from aliphatic precursors, as the epitaph of the empirical isoprene rule. The biogenetic isoprene rule was enunciated as a comprehensive constitutional theory based on reaction mechanism. Different structural types of cyclic mono-, sesqui-, di- and triterpenes were to be derived from acyclic precursors via cationic addition, cyclization, and 1,2-rearrangement reactions (see Fig. 17). The central paragraph reads as follows:

«...the deduction of structures of natural terpenic compounds by accepted reaction mechanisms from the hypothesized single precursors squalene, geranylgeraniol, farnesol, geraniol ("biogenetic isoprene rule") not only serves to outline possible biogenetic routes, but also represents a new helpful tool

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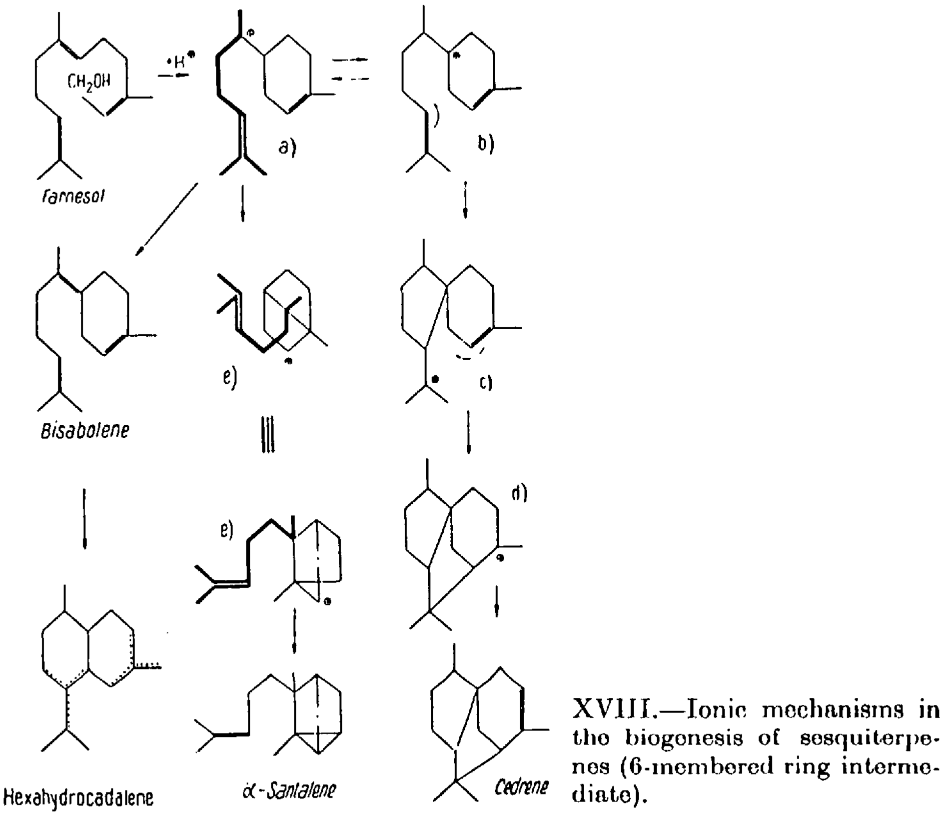
[11*] In a lecture before the Swiss Chemical Society in Zürich in summer 1923 on bile acids, *Heinrich Wieland* had raised the possibility that these were biogenetically derived from lipids. In the ensuing discussion *Ruzicka* pointed to a possible common origin for sterols and higher terpenes. This hypothesis is described in the dissertation of *Rudolph*[82], one of *Ruzicka*'s doctoral students, which appeared in printed form in 1925; cf. also[5,9].



XVIII.—Ionic mechanisms in the biogenesis of sesquiterpenes (6-membered ring intermediate).

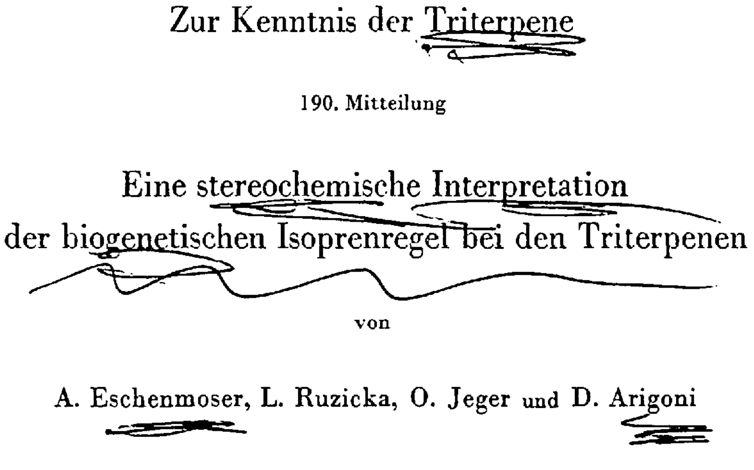
Fig. 17. Excerpt from «The Isoprene Rule and the Biogenesis of Terpenic Compounds» [5], *1953 (cf. also Fig. 19).*

in the structure elucidation of terpenic compounds. The new tool limits the number of carbon skeletons which can be proposed on the basis of the original isoprene rule ("empirical isoprene rule") alone.»

Impetus and background for the biogenetic isoprene rule paper were provided by *Jeger*'s and *Ruzicka*'s lanosterol work, the *Woodward-Bloch* proposal, and the author's concepts of deriving the constitutional formulae of terpenes by coherent schemes involving cationic cyclizations and rearrangements [13*]: complex sesquiterpenes (cedrene, caryophyllene) from farnesene [64] [12*]; abietic acid and related diterpenes from the naturally occurring precursor manool [64]; and, dating from 1953, all triterpenes of then known constitution from squalene. The derivation of the triterpene formulae was based on the new concept of the *oxidative* initiation of cyclization of terpenoid polyene chains [64]. In this the hydroxy group in ring A was to be seen as a crucial functional group, and its origin a potential hydroxyl cation as cyclization initiator. This latter concept inspired *Bloch*'s experiments on the role of atmospheric oxygen in cholesterol biosynthesis [86] (cf. [83, 87]) and some years later the search for squalene oxide by *Corey* [88] and by *van Tamelen* [89]. A final impetus was provided by the tricyclic triterpene alcohol ambrein, whose structure was solved in 1947 by *Lederer* [90] working in competition with the ETH group [91]. This structure was clearly to be derived by squalene cyclization (this time by a non-oxidative mechanism).

Admittedly *Ruzicka*'s 1953 paper was decisively influenced and abetted by the work of others, but only he was capable and authoritative enough to give it the standing of a manifesto which was to receive general recognition as signifying the changes wrought in terpene chemistry by the new concepts. The biogenetic isoprene rule would now make obsolete all the old structural distinctions previously made on the basis of either regular or irregular isoprene array, that is, of obeying or not obeying the old rule. This transition is surely among the most beautiful examples of the evolution of a chemical theory. Perhaps the profoundest aspect of the new rule was as a further step forward in the demystification of the origin of natural products which began with *Wöhler*'s 1828 urea synthesis. It served to demolish the last vestige of the myth of a «Force Vitale» – this time in the form of a belief that the enzymatic course of biosynthesis can go by way of reactions of a type unknown to the chemist. In this connection it is historically wrong to make light of (mechanistically) the squalene folding concept put forward by *Robinson* in 1934 [84] (something which the author of these lines has himself been guilty of). The Robinson concept should be looked at as an historically important document, describing as it does the prevalent view at that time on the then unknown territory of biosynthesis, and that against the background of his standing in mechanistic organic chemistry at the time. His approach to the isoprene rule (see Fig. 10) is a similar document.

The immediate universal acceptance accorded to the biogenetic isoprene rule was clearly due to the way it managed at a stroke to put order among the overwhelmingly large number of cyclic terpenes which had become known during the preceding 70 years – this on the basis of just a few organic reaction concepts. Later, in 1955, it became all the more convincing in the way it was instrumental in deriving not only the constitution but the *configuration* of all known triterpenes derived from squalene, via definition of the steric course


Zur Kenntnis der Triterpene

190. Mitteilung

Eine stereochemische Interpretation
der biogenetischen Isoprenregel bei den Triterpenen

von

A. Eschenmoser, L. Ruzicka, O. Jeger und D. Arigoni

Fig. 18. Ruzicka's personal copy of the 1955 paper [6].

[12*] These reaction schemes were published in detail for the first time in [5]; see e.g. Fig. 17.

[13*] According to the author's recollection the «rate-limiting step» in devising this scheme lay in overcoming the «activation barrier» represented by having to disregard *Markovnikov*'s rule where the cyclization to ring C of the triterpenes was concerned. Had it not been for this, the whole scheme might have come to fruition three years earlier. It was *H. Heusser* (*K. Bloch* was then a guest in his laboratory) who first placed before *Ruzicka* a scheme which represented an attempt to derive triterpenes from squalene. This scheme was quite different from that presented in [5], but, as far as the author remembers, did include anti-Markovnikov cyclizations; it is quite likely that this helped in overcoming the «activation barrier».

of acid-catalyzed polyene cyclizations and Wagner-Meerwein shifts. In later years[92] *Ruzicka* indicated that it was he who had induced his younger colleagues (*A. E.* and *Jeger*'s rising star student *Duilio Arigoni*) to solve the stereochemical aspects of the problem. Be that as it may, solved it was indeed by these younger colleagues, and the result was a publication in 1955 in Helvetica Chimica Acta[6] which was greeted as «the apotheosis of the isoprene rule»[94] by *John W. Cornforth*, a competent witness on the strength of his fundamentally important contributions to the study of the biosynthesis of cholesterol[93]. Among the papers left by *Ruzicka* we can find a reprint of this article with parts underlined by him (see Fig. 18).

The essence of this development was the establishment of two principles: one, on the preferential stereoelectronic course of electrophilic addition and substitution reactions at a C=C double bond, in conjunction with accompanying or subsequent Wagner-Meerwein rearrangement, the other the conformational one based on the dichotomy of chair versus boat form of the polyene chain. For those who were involved in this work it was an extraordinary experience to follow up how different permutations of constitutional and conformational variation in the squalene molecule could now be shown to lead to the complete structure and shape of any one of all the known triterpenes, all on the basis of an amazingly small number of strictly defined reaction postulates.

Things were again developing rapidly: at Columbia University *Stork*, independently of the author's work at the ETH[6,95,96], had initiated experiments on the steric course of polyene cyclization using a working hypothesis of his own and had moved toward a rationalization of the biogenetic path from squalene to the triterpenes[97]. At Harvard University *Woodward* had (also independently) lectured[98] during the winter semester 1954/55 on the constitutional and configurational derivation of lanosterol and euphol from squalene, likewise using the argument of chair versus boat conformation of the squalene chain.

As a theory covering both molecular architecture and natural origin of the terpenes the biogenetic isoprene rule has since its inception in 1953–1955 changed the course of study of these natural products. The number of new sesqui-, di- and triterpenes since studied and found to be new variants within the original concept has become virtually limitless (at least so it seems to this author). As a result the rule has become instrumental in the metamorphosis of natural product chemistry into bioorganic chemistry, and in forging the link between organic chemistry and biology.

Initially the rule had been mainly concerned with the cyclic terpenes. The scope changed in the course of the discovery of acetyl coenzyme A by *Lynen*[99] in 1951, that of mevalonic acid by *Folkers* in 1956[100], and the subsequent work of *Lynen*[101], *Bloch*[102], and of *Cornforth* and *Popjak*[103], which all resulted in the complete picture of the biosynthesis of squalene starting from acetic acid. An important «fall-out» of all this was the recognition of isopentenyl pyrophosphate as the true incarnation of the legendary «isoprene unit», thus laying the biochemical foundation of the biogenetic isoprene rule. Further subsequent tests of its validity were the continuing work on the biosynthesis of cholesterol (cf.[88,89,104,105]), and the incisive investigations of selected cyclic terpenes, mainly by *Arigoni* (cf.[106]) and his school[107].

Fig. 19 reproduces a selection of handwritten notes made by *Ruzicka* for his lecture at the Nobel Laureates' meeting in Lindau in 1967[2]. They give us an idea of how he himself viewed all these developments. The photograph in Fig. 20 showing *Ruzicka* and *Lynen* together most probably also dates from that meeting.

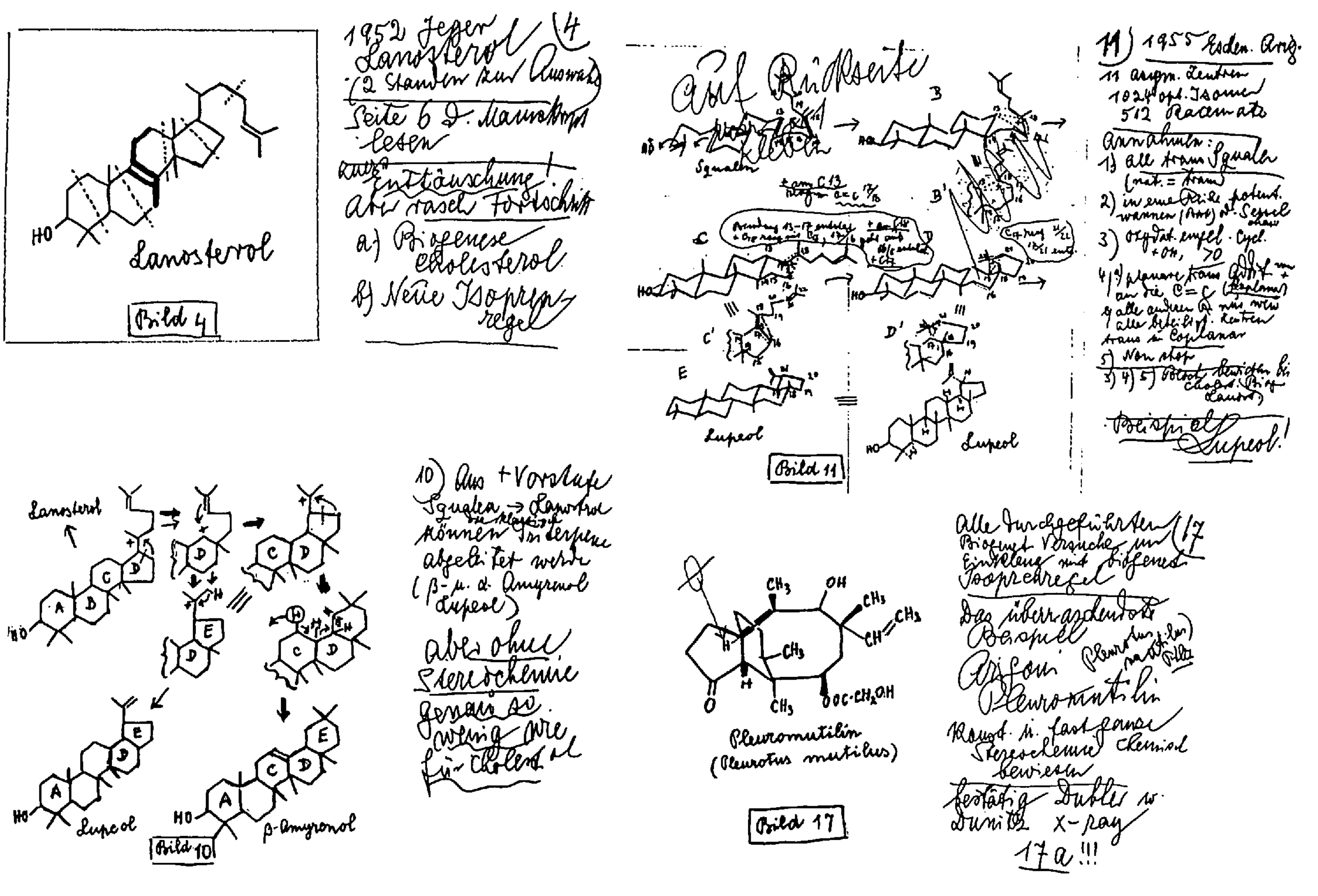


Fig. 19. Excerpts from manuscript drafts for a lecture by Ruzicka on the biogenetic isoprene rule[2].

Fig. 20. *Leopold Ruzicka (with Feodor Lynen in the background) at the meeting of Nobel laureates in Lindau (probably 1967); Photo-Pfeiffer, D–7993 Kressbronn.*

In 1956 *Ruzicka* contributed his article «The Significance of Theoretical Organic Chemistry for the Chemistry of the Terpenes»[108] to «Perspectives in Organic Chemistry», *Todd*'s valedictory volume for *Robert Robinson.* In this he surveyed the entire story, from *Wallach*'s «Terpenes and Camphor»[17] to the stereochemical implications of the biogenetic isoprene rule; from his early 1918 work on Wagner rearrangements in the pinene series[30] via the views of *Meerwein* and *Winstein* on the nature of carbocation reactions, to the latest experimental results on the course of acid-catalyzed polyene cyclization. This was the new *Ruzicka* at the height of his powers. He was in no way less enthusiastic about reaction mechanisms and biosynthesis than his junior colleagues who were assisting him. At the end of the article we find the following observations:

«The enormous progress we have seen in elucidation of structure since the days of Wallach is evident from the way in which the minutest detail in the structures of the steroids and triterpenes can be accounted for by the principles of the biogenetic isoprene rule. For the way the true picture has emerged from behind the facade of the structural formula we have to thank modern developments in reaction mechanism – a triumph over the rigid system of mere structural concepts. No other group of natural products has so far lent itself to the same extent to rationalization of structural and configurational detail by simplest means as that of the triterpenes and steroids. The ability of so many living organisms to biosynthesize all these compounds with always the same configuration, out of hundreds of alternatives, must surely be one of the most remarkable chemical achievements of Nature.»

Ruzicka was fortunate in witnessing the consequences and approbation of his success for many more years of his life. After his formal retirement in 1957 he continued giving lectures to international audiences, notably his famous Faraday Lecture[27], «History of the Isoprene Rule», which particularly contributed to the propagation of the new concepts. Reading once again that lecture, as well as many unpublished manuscripts of other lectures (cf. Fig. 19), one cannot but sense the feeling of satisfaction experienced by *Ruzicka* in his seventies and eighties, at his extraordinary good fortune in seeing the essence of his life's work, the isoprene rule, coming to final fruition.

The Turn to Biochemistry

The encounter between natural products chemistry and biochemistry, within the arena provided by the advent of the study of lanosterol, was the high point of terpene chemistry in the 1950's. For *Ruzicka,* who set the stage, it signified a common future course for both disciplines, and he lost no time in translating his convictions into concrete action. This was to take the form of establishing an Institute of Biochemistry at the ETH and of raising the necessary financial support by an appeal to chemical industry in Basel. Moreover, he personally took in hand the creation of a Chair in Biochemistry, and for him the first choice would have had to be between *Konrad Bloch, Feodor Lynen,* and *George Popjak* – at that time the leading figures in the field of cholesterol and terpenoid biosynthesis. Concurrently, and well into the sixties, *Ruzicka* made an extraordinary effort to familiarize himself with the biochemical literature. It is clear from the notes and sketches left after his death that, had he been able to complete his autobiography, it would have been under the heading «My Predilection for the Chemistry of Life»[109]. The following typifies his intense preoccupation with that subject:

«I would define as "Chemistry of Life" the collaboration between the chemistry of organic natural products (= bio-organic chemistry) and biochemistry. The task of bio-organic chemistry is to elucidate in the laboratory the structures and reactions of naturally occurring organic compounds, whereas that of biochemistry is to study and clarify the course of chemical reactions within living organisms» [3].

For *Ruzicka* natural product chemistry had always represented an essential prerequisite for biochemistry. He had none of the misgivings that presently underlie the debate on the relationship between organic chemistry and biochemistry. In fact, he could express himself on that subject in no uncertain terms[3]:

«There are some small-minded organic chemists who on occasion bewail the fact that a good part of organic chemistry has been expropriated by biochemistry. They have forgotten that organic chemistry originally set out to clarify the chemistry of car-

bon compounds found in living organisms. It is the level of perfection achieved by organic chemistry after 150 years which is enabling biochemistry to penetrate the innermost secrets of life processes on a molecular basis, and this will more and more blur the distinction between the two disciplines. It brings to mind my favourite paradox: "To understand biochemistry you need to know at least as much organic chemistry as for organic chemistry itself".»

Ruzicka's convictions on this subject are eloquently clear both from the title of his last autobiographical publication («In the Borderland between Bioorganic and Biochemistry») as well as from its venue of appearance (Annual Reviews of Biochemistry 1973[9]).

Among the many drafts of lectures left by him, one stands out in particular. It is of a talk under the heading «Biochemie und Weltbild»[3] which *Ruzicka* gave at the University of Bern in 1964 and for which further dissemination had been intended, both as a radio talk as well as by a separate publication[110]. On one copy of the manuscript we find a hand-written list of headings which it is pertinent to reproduce here because of the way it illustrates the broad range of his interests and of his familiarity with the biochemical literature of that period. After first defining the mission of biochemistry (see above quotation), briefly outlining its history and explaining the meaning of terms such as protein and enzyme[14*], he proceeds with the following topics (examples shown in Fig. 21 on p. 18/19):

«5. Dependence of biological activity on constitution[14*]
6. Genetics and biochemistry
7. Nucleic acids
9. Duplication of DNA[14*]
11. Biological synthesis of RNA in vitro[14*]
12. DNA-directed protein synthesis
  (a) Historical introduction
  (b) Messenger RNA
  (c) Transfer- (or) s-RNA
  (d) Code
13. Determination of code triplets
14. Mutation and inheritance
15. Mutation and evolution, cytochrome c
16. Biochemical evolution
17. Significance of biochemistry in evolution
18. Did evolution have specific aims?
19. Problems of the future
20. Biochemistry of the brain
21. Organic and biochemistry»

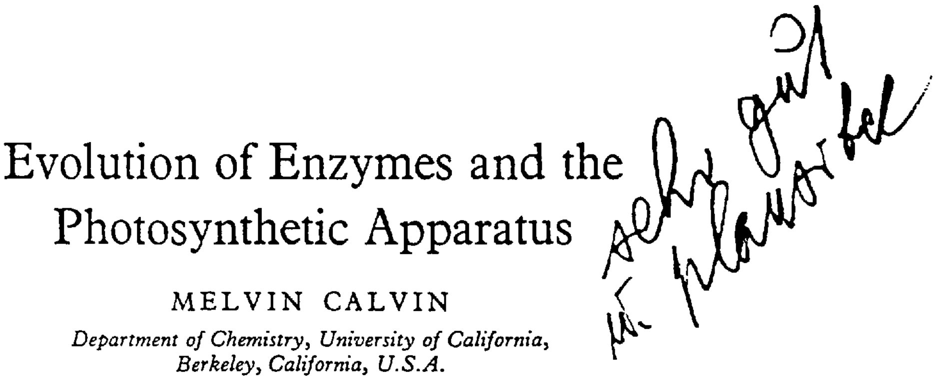
This, and the text of the lecture itself in its clarity and precision, are vivid evidence of the vitality, at the age of 77, of a man of broad vision, with remarkably comprehensive awareness of the central themes of the new discipline of molecular biology.

1953 was not only the year of the biogenetic isoprene rule; it also marked the advent of the structure model of DNA by *Watson* and *Crick*[111]. We can well imagine *Ruzicka*'s excitement during the following years at the unexpected and extraordinary developments taking place in the realm of natural products outside the traditional territory of purely organic chemistry. If we accept his above-mentioned definition of bio-organic chemistry at face value – and in the long run it may well be more valid than the current version – then it would certainly encompass a discovery such as that of the double helix structure of DNA. We organic natural product chemists should keep in mind *Ruzicka* and his definition whenever and wherever the project on sequencing the human gene, i.e. «the determination of constitution of the natural product human DNA» comes up.

1953 was also the year of the *Miller* experiment which first conclusively demonstrated the formation of alpha-amino acids, building blocks of the proteins, by electric discharge through mixtures of methane, ammonia, hydrogen, and water[112]. This was to be the classical experiment laying an experimental foundation for prebiotic chemistry, a subject first propounded in 1924 by the Russian biochemist *Oparin* through his hypothesis on the origin of life on earth[113].

Questions on the Origin

The «First International Symposium on the Origin of Life on Earth»[114], organized by *Oparin*, took place in Moscow in 1957; its Proceedings appeared in print two years later. The 500 or so scientists who took part included a number of prominent chemists of varied background, such as *Pauling, Calvin, Stanley, Prigogine, Bernal, Mothes, Chargaff* and others. *Ruzicka*'s personal copy of the Proceedings provides evidence of his fascination with this meeting; his hand-written comments cover the list of contents and abstracts of lectures by, e.g., *Urey* (primordial atmosphere), *Miller* (experiments on prebiotic chemistry), and *Stanley* (nature of viruses). Certain authors were the object of forthright comments (Fig. 22), such as *Stanley* (*"Bravo"*, see Fig. 23), or *Chargaff*, whose already then evident skepticism of the scientific ap-



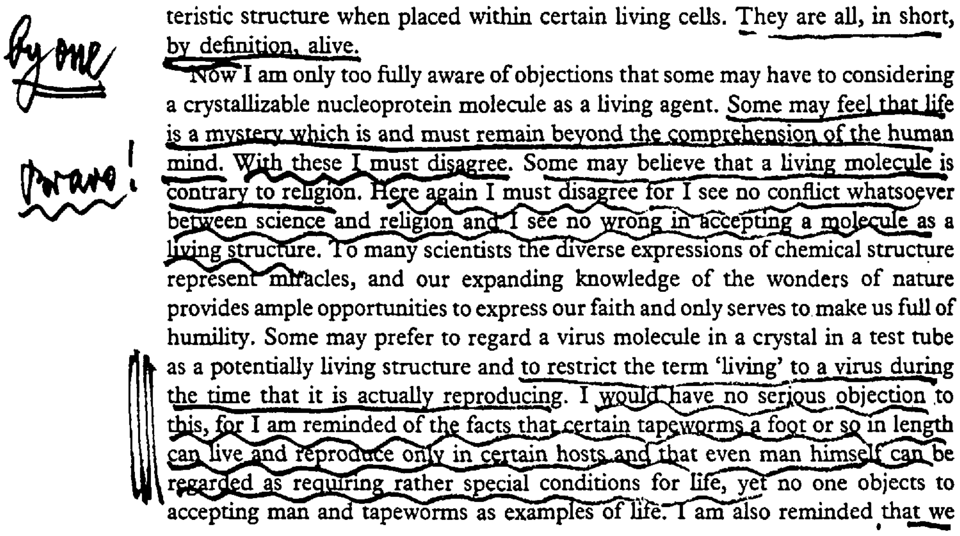
Evolution of Enzymes and the
Photosynthetic Apparatus

MELVIN CALVIN

Department of Chemistry, University of California,
Berkeley, California, U.S.A.

sehr gut
u. [illegible]

Fig. 22. Excerpts from[114].



by one

Bravo!

teristic structure when placed within certain living cells. They are all, in short, by definition, alive.

Now I am only too fully aware of objections that some may have to considering a crystallizable nucleoprotein molecule as a living agent. Some may feel that life is a mystery which is and must remain beyond the comprehension of the human mind. With these I must disagree. Some may believe that a living molecule is contrary to religion. Here again I must disagree for I see no conflict whatsoever between science and religion and I see no wrong in accepting a molecule as a living structure. To many scientists the diverse expressions of chemical structure represent miracles, and our expanding knowledge of the wonders of nature provides ample opportunities to express our faith and only serves to make us full of humility. Some may prefer to regard a virus molecule in a crystal in a test tube as a potentially living structure and to restrict the term 'living' to a virus during the time that it is actually reproducing. I would have no serious objection to this, for I am reminded of the facts that certain tapeworms a foot or so in length can live and reproduce only in certain hosts and that even man himself can be regarded as requiring rather special conditions for life, yet no one objects to accepting man and tapeworms as examples of life. I am also reminded that we

Fig. 23. Excerpt from[114]*; Ruzicka's marginal notes on W. M. Stanley's paper «On the Nature of Viruses, Genes, and Life».*

[14*] The first three pages, as well as pages 6 and 10 of the copy of this manuscript are missing.

proach drew the terse observation:

«Ch. commits the opposite mistake to that of the Origin-of-Life scribblers» (Fig. 24)

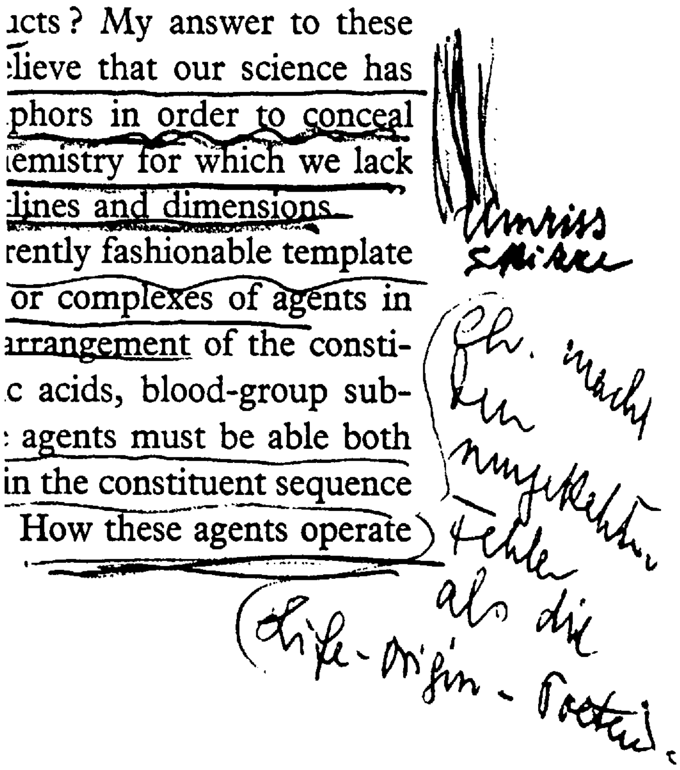
a judgment illuminative not only of *Chargaff* but of *Ruzicka* himself – his both open and critical attitude to the origin-of-life issue.

It was not entirely surprising that *Ruzicka* should have begun in his later years to evince interest in the origin-of-life problem. The search for the origin of biosynthetic pathways was a logical outcome of his endeavours to gain an insight into the molecular construction principles of nature: first on the level of organic natural products chemistry and then on the bioorganic level of biosynthesis. His life-long fascination with the topic «Chemistry of Life» led naturally to his preoccupation with that of the «Origin of Life». Presumably he agreed with *Oparin*'s postulate that «the real nature of life is comprehensible only through understanding of its origin» ([114], p. 368).

In the manuscript of his «Biochemie und Weltbild» lecture[3] *Ruzicka* devoted nearly 3 out of 23 pages to the subject of the origin of life and evolution, but struck a rather cautious note, perhaps in view of the nature of his audience. (Bio)chemical evolution[15*], in his view, ought to be considered an essential prerequisite of biological evolution, the latter being now almost universally accepted; even its opponents could hardly object to the idea of (bio)chemical evolution since it would apply only to the creation of inanimate organic chemical compounds. He then wrote:

> *«The crux of the concept of evolution as a whole lies in the fact that somewhere in between (bio)chemical and biological evolution we must assume a point where life was created no matter whether its origin is regarded as "natural" or "supernatural"»* [3].

We should not overlook the fact that eventually *Ruzicka* decided to strike out the end of the last sentence (see Fig. 21). That act was in line with innumerable notes and comments from the 1960's which leave no doubt that from both a scientific and emotional viewpoint he considered the creation of life as a consequence of the intrinsic properties or organic matter on earth. His perusal of the publications of experimentally active protagonists of prebiotic chemistry such as *Calvin*[115,116], *Miller*[117], and *Ponnamperuma*[118] is littered by marginal notes expressing emphatic approval (*"very good", "very interesting and important", "excellent"*). Comments in a different vein adorn his copy of «Life transcending Physics and Chemistry»[119], a significant article by the physical chemist and philosopher *Michael Polanyi*. For *Ruzicka*, organic chemistry was the epicenter of his private universe; his approval of other authors' work was in


ıcts? My answer to these
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c acids, blood-group sub-
: agents must be able both
in the constituent sequence
How these agents operate

Umriss Skizze

Ch. macht umgekehrte Fehler als die (Life-Origin-Poeten)

Fig. 24. Excerpt from [114]; Ruzicka's marginal notes on E. Chargaff's paper «Nucleic Acids as Carriers of Biological Information».

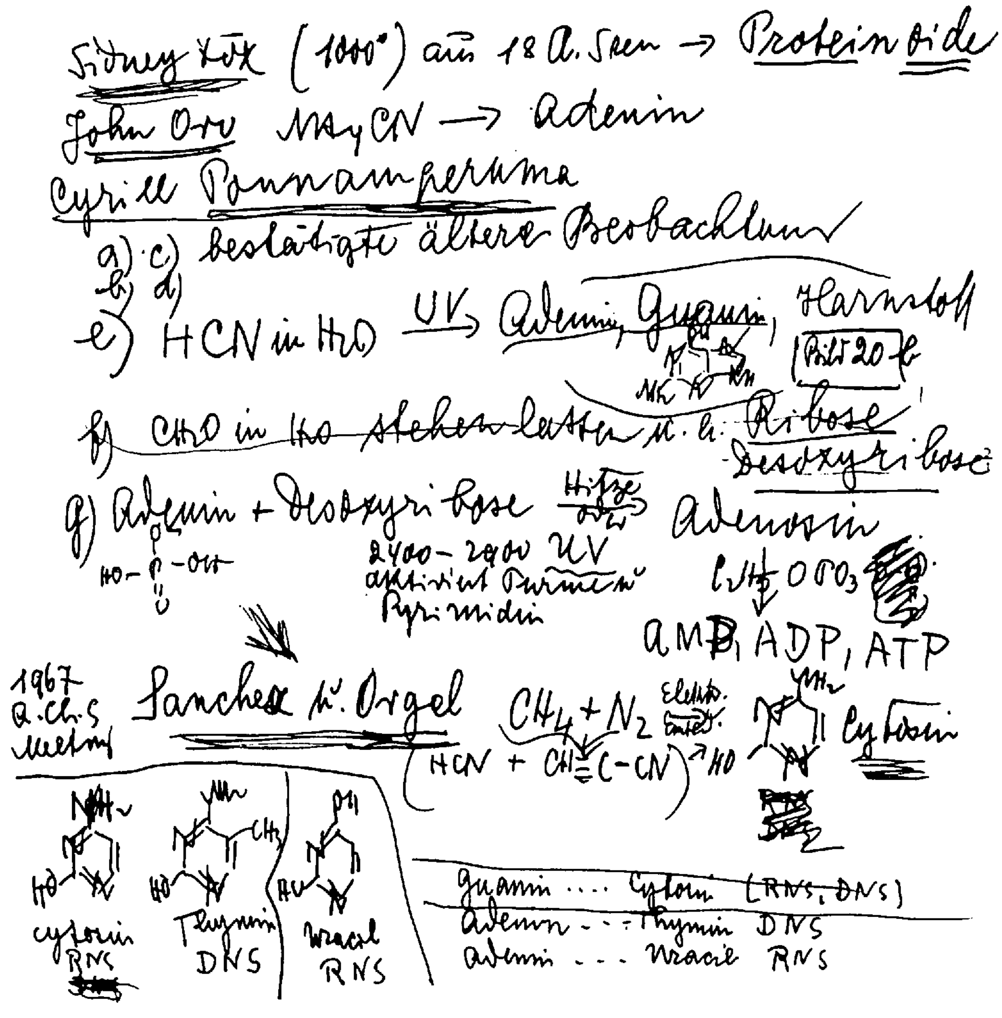
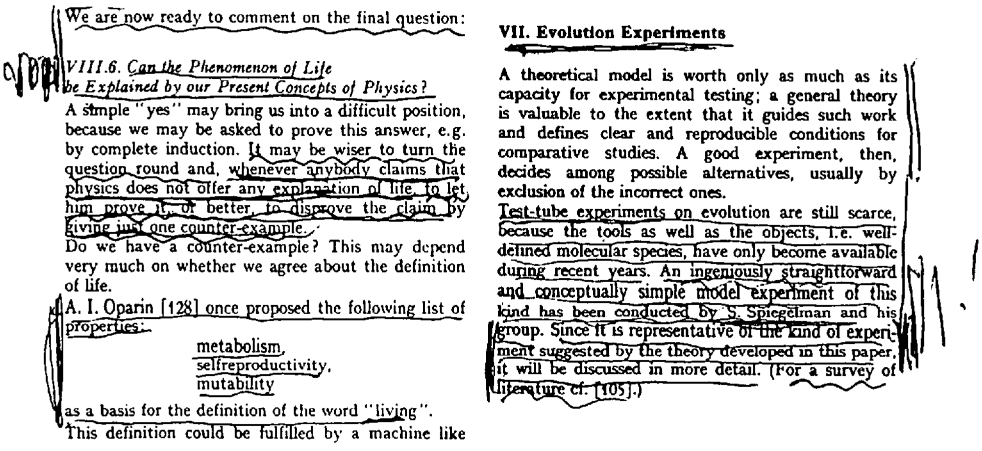


Fig. 25. Ruzicka's sketches on the subject of «prebiotic chemistry».

[15*] *Ruzicka* here used the term «biochemical evolution» in the sense of *Calvin*'s use of «chemical evolution»[115].

proportion to the extent to which it would correlate the structures and properties of organic compounds with their biological significance. For him as an organic chemist the discipline of prebiotic chemistry was closest to his heart, judging by the way he systematically kept himself up-to-date in this field by hand-written excerpts from Annual Reports on the Progress of Chemistry 1967–1968 (see Fig. 25).

None of this quite accounts for the way *Ruzicka*, then already in his eighties, was fascinated by the young *Manfred Eigen*. We must marvel at *Ruzicka*'s attempts to come to terms with a theory of evolution which was largely based on mathematical principles. From his hand-written excerpts from *Eigen*'s Nobel lecture on «Immeasurably Fast Reactions» we can gather how much he was impressed by *Eigen*'s brilliance of exposition well beyond the narrow confines of chemistry. One of the important items among the *Ruzicka* papers is the issue of Naturwissenschaften from the year 1971 with *Eigen*'s article: «Self Organization of Matter and the Evolution of Biological Macromolecules»[120], annotated by *Ruzicka*. In retrospect this is a touching token of an 84-year old *Ruzicka* still determined to add to his fund of knowledge and understanding of the «Chemistry of Life» (cf. Fig. 26).



We are now ready to comment on the final question:

VIII.6. Can the Phenomenon of Life be Explained by our Present Concepts of Physics?

A simple "yes" may bring us into a difficult position, because we may be asked to prove this answer, e.g. by complete induction. It may be wiser to turn the question round and, whenever anybody claims that physics does not offer any explanation of life, to let him prove it, or better, to disprove the claim by giving just one counter-example.

Do we have a counter-example? This may depend very much on whether we agree about the definition of life.

A. I. Oparin [128] once proposed the following list of properties:

metabolism,
selfreproductivity,
mutability

as a basis for the definition of the word "living".

This definition could be fulfilled by a machine like

VII. Evolution Experiments

A theoretical model is worth only as much as its capacity for experimental testing; a general theory is valuable to the extent that it guides such work and defines clear and reproducible conditions for comparative studies. A good experiment, then, decides among possible alternatives, usually by exclusion of the incorrect ones.

Test-tube experiments on evolution are still scarce, because the tools as well as the objects, i.e. well-defined molecular species, have only become available during recent years. An ingeniously straightforward and conceptually simple model experiment of this kind has been conducted by S. Spiegelman and his group. Since it is representative of the kind of experiment suggested by the theory developed in this paper, it will be discussed in more detail. (For a survey of literature cf. [105].)

Fig. 26. Excerpts from Ruzicka's copy of the journal Naturwissenschaften containing Eigen's publication[120].

What was the effect *Eigen* eventually had on *Ruzicka*? An answer is provided by an incident which occurred at the 1971 Bürgenstock conference[121]. *Eigen* had just finished giving his lecture on «Evolution of Biological Macromolecules», whereupon an agitated *Ruzicka* stood up and said:

«It has taken all my life's work to convince myself that life is chemistry; and now you come along and tell us it is physics.»

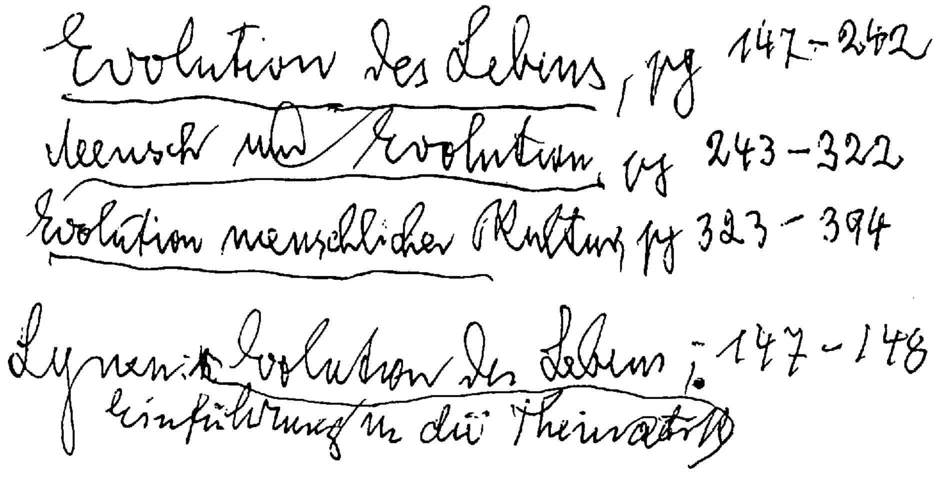
Apart from causing much amusement among the audience, this rejoinder went right to the heart of the matter. At a time when experimental endeavour in the area of the origin-of-life problem was overshadowed by the brilliance of physical theories – the kinetic concepts of evolution by *Eigen*[120,122] and the thermodynamic perceptions of *Prigogine*[123] – it was fitting that *Ruzicka* the chemist should intuitively have made a point that today has moved to the forefront of interest: the problem of the origin of life on earth is above all a chemical one, and specifically also a problem of synthetic organic chemistry. A chemical synthesis proceeds within the confines of kinetic and thermodynamic principles; but a mere description of these cannot by itself describe that synthesis. Description of a chemical synthesis is first of all a delineation of its course in terms of molecular structure; and there is no reason why this should not apply equally to a theory of the origin of life on earth[124].

Material testifying to *Ruzicka*'s concern with questions of evolution can be found up to his 89th year of life. This includes, e.g., marginal comments which appear in the 1975 Symposium Volume on «Evolution» issued by the Akademie der Deutschen Naturforscher Leopoldina (cf. Fig. 27), specifically on *Kuhn*'s essay «On the Evolution of a Self-organizing Prebiotic System»[125,126].

Among the many loose sheets found among the *Ruzicka* papers there is one on which he constructed a sentence around an important concept in the history of science – «Force Vitale» – which is a valid reinterpretation of that concept and at the same time an enunciation of *Ruzicka*'s scientific credo:

«The Life Force, abused so often in days gone by as a deus ex machina, is not dethroned: it is as mighty as ever; but the irrevocable evolution of the sciences reveals that force to be molecular and physicochemical in essence» (Fig. 28).

Ruzicka's work on the isoprene rule indeed contributed to that revelation.



Evolution des Lebens, pg 147-242
Mensch und Evolution, pg 243-322
Evolution menschlicher Kultur pg 323-394

Lynen: Evolution des Lebens; 147-148
Einführung in die Thematik

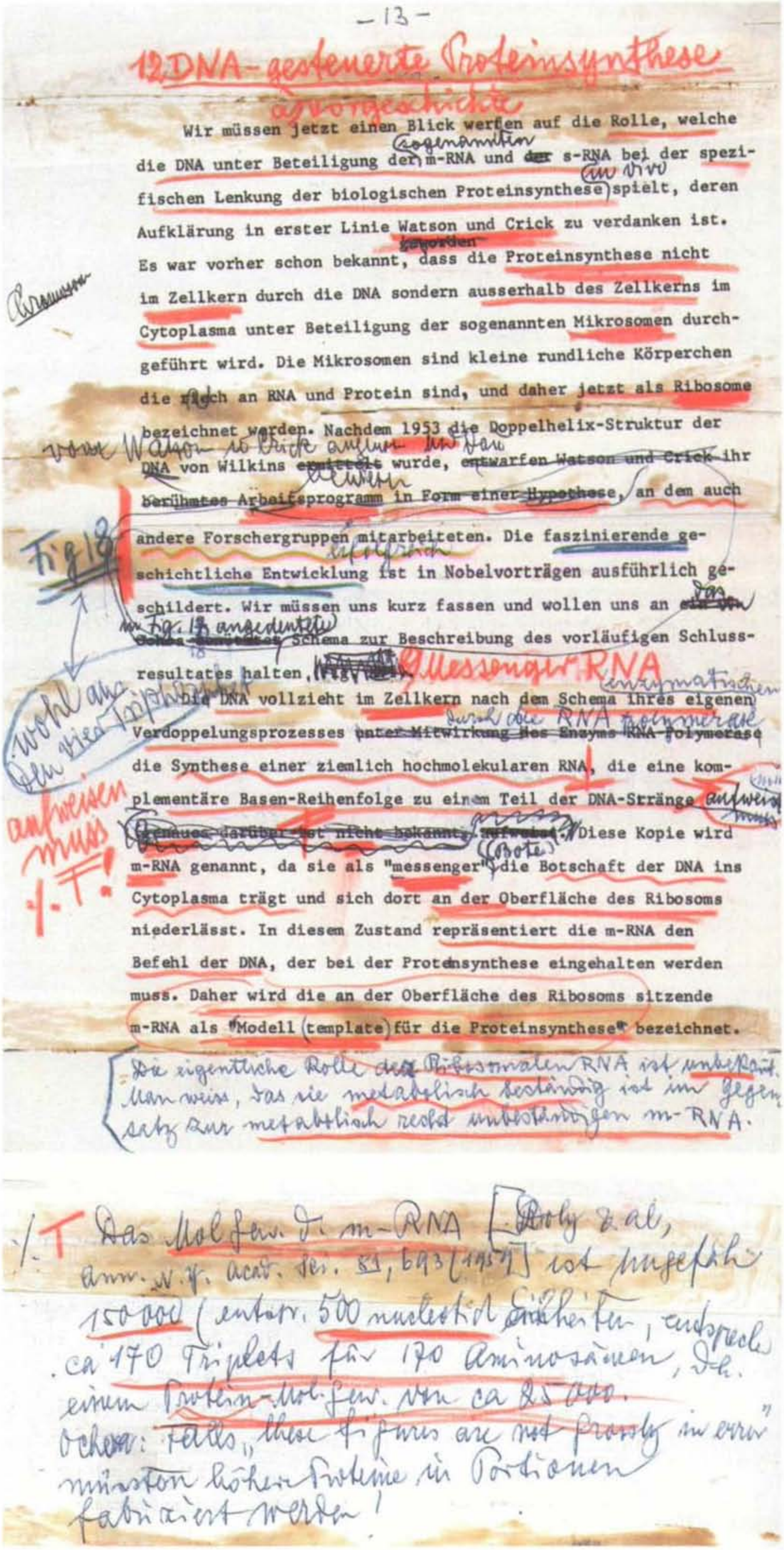
Fig. 27. Ruzicka's handwriting, 1975 or 1976[125].

Postscript

It was on the occasion of the Ruzicka Centennial Symposium in Zürich that the author agreed to accede to Professor *K. Balenović*'s request to write an article about *Ruzicka*. What I had in mind was a short essay on the theme «Leopold Ruzicka and the problem of the origin of life». What came about in the end, as a result of closer preoccupation with that subject and with the *Ruzicka* papers, was an attempt to present this theme in conjuction with an account of the history of the isoprene rule. In particular it also resulted in an account of the origins of the biogenetic isoprene rule, from the special viewpoint of the author who saw its beginnings, and believes that after 35 years he has possibly attained a balanced perspective.

What seems certain is that whoever will chance to acquaint himself with *Ruzicka*'s scientific legacy in the future cannot but come under the spell of his extraordinary personality, and will regret, as the author does, that it was not to be in his power to complete his autobiography.

Fig. 21. Reproduction of selected pages from the manuscript « Biochemie und Weltbild » [3].



-13-

12 DNA-gesteuerte Proteinsynthese

a) Vorgeschichte

Wir müssen jetzt einen Blick werfen auf die Rolle, welche die DNA unter Beteiligung der sogenannten m-RNA und ~~der~~ s-RNA bei der spezifischen Lenkung der biologischen Proteinsynthese in vivo spielt, deren Aufklärung in erster Linie Watson und Crick zu verdanken ist. Es war vorher schon bekannt geworden, dass die Proteinsynthese nicht im Zellkern durch die DNA sondern ausserhalb des Zellkerns im Cytoplasma unter Beteiligung der sogenannten Mikrosomen durchgeführt wird. Die Mikrosomen sind kleine rundliche Körperchen die reich an RNA und Protein sind, und daher jetzt als Ribosome bezeichnet werden. Nachdem 1953 die Doppelhelix-Struktur der DNA von Watson u. Crick anhand der Daten von Wilkins ~~ermittelt~~ erkannt wurde, ~~entwarfen Watson und Crick ihr berühmtes Arbeitsprogramm in Form einer Hypothese~~, an dem auch andere Forschergruppen erfolgreich mitarbeiteten. Die faszinierende geschichtliche Entwicklung ist in Nobelvorträgen ausführlich geschildert. Wir müssen uns kurz fassen und wollen uns an ~~ein von~~ das in Fig. 18 angedeutete ~~Crick skizziertes~~ Schema zur Beschreibung des vorläufigen Schlussresultates halten, ~~(Fig. 18)~~.

Chromosom

Fig 18

b) Messenger-RNA

Die DNA vollzieht im Zellkern nach dem Schema ihres eigenen enzymatischen Verdoppelungsprozesses durch die RNA Polymerase ~~unter Mitwirkung des Enzyms RNA-Polymerase~~ die Synthese einer ziemlich hochmolekularen RNA, die eine komplementäre Basen-Reihenfolge zu einem Teil der DNA-Stränge aufweist ~~Genaueres darüber ist nicht bekannt, aufweist~~. Diese Kopie wird m-RNA genannt, da sie als "messenger" (Bote) die Botschaft der DNA ins Cytoplasma trägt und sich dort an der Oberfläche des Ribosoms niederlässt. In diesem Zustand repräsentiert die m-RNA den Befehl der DNA, der bei der Proteinsynthese eingehalten werden muss. Daher wird die an der Oberfläche des Ribosoms sitzende m-RNA als "Modell (template) für die Proteinsynthese" bezeichnet.

wohl aus den vier Triphosphaten

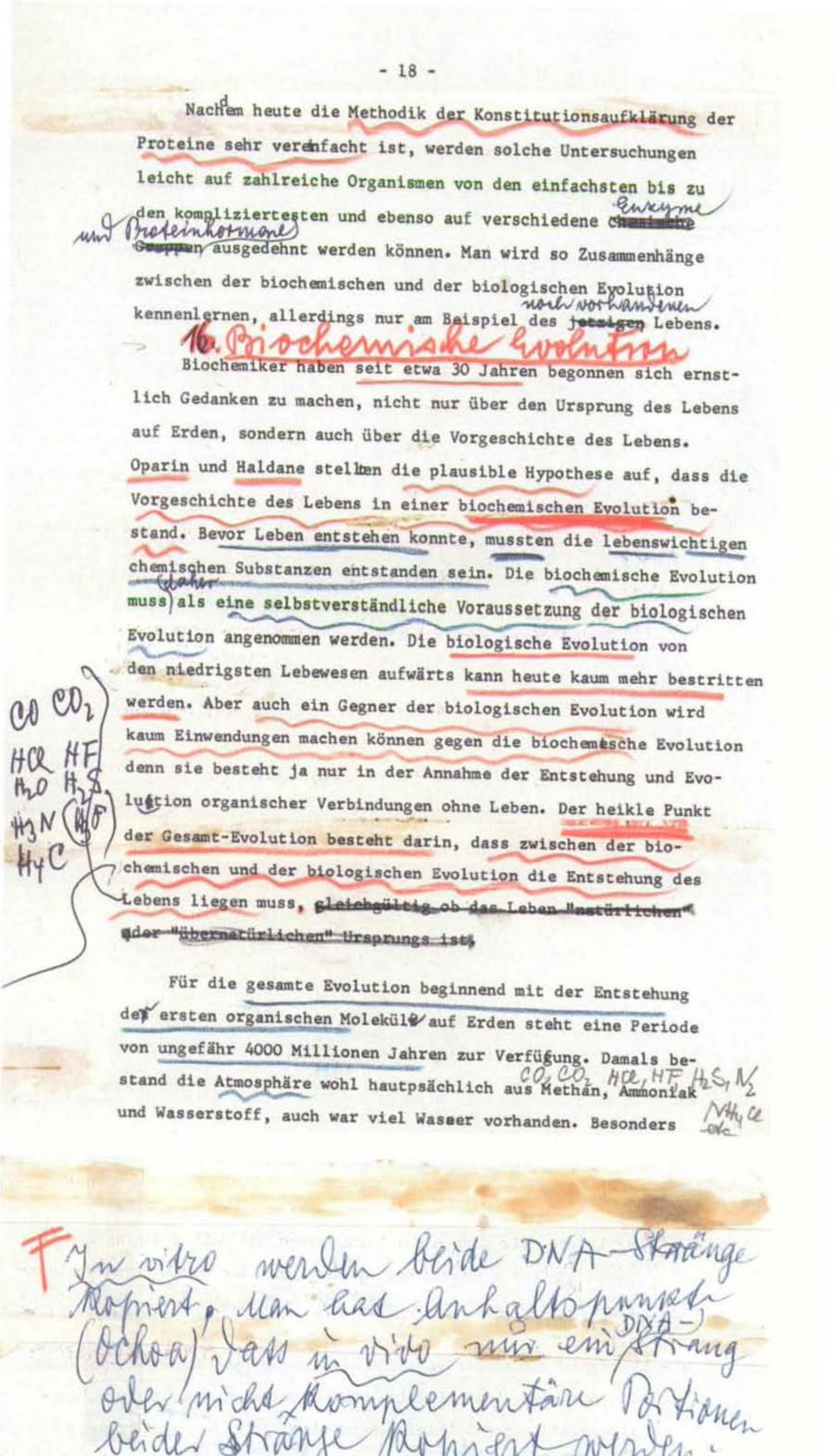
aufweisen muss

1-T!

Die eigentliche Rolle der ribosomalen RNA ist unbekannt. Man weiss, das sie metabolisch beständig ist im Gegensatz zur metabolisch recht unbeständigen m-RNA.

1-T Das Mol.Gew. d. m-RNA [Doty et al, Ann. N.Y. Acad. Sci. 81, 693 (1959)] ist ungefähr 150 000 (entspr. 500 nucleotid Einheiten, entsprechend ca 170 Triplets für 170 Aminosäuren, d.h. einem Protein-Mol.Gew. von ca 25 000.

Ochoa: Falls „these figures are not grossly in error" müssten höhere Proteine in Portionen fabriziert werden!



- 18 -

Nachdem heute die Methodik der Konstitutionsaufklärung der Proteine sehr vereinfacht ist, werden solche Untersuchungen leicht auf zahlreiche Organismen von den einfachsten bis zu den kompliziertesten und ebenso auf verschiedene ~~chemische Gruppen~~ Enzyme und Proteinhormone ausgedehnt werden können. Man wird so Zusammenhänge zwischen der biochemischen und der biologischen Evolution kennenlernen, allerdings nur am Beispiel des ~~jetzigen~~ noch vorhandenen Lebens.

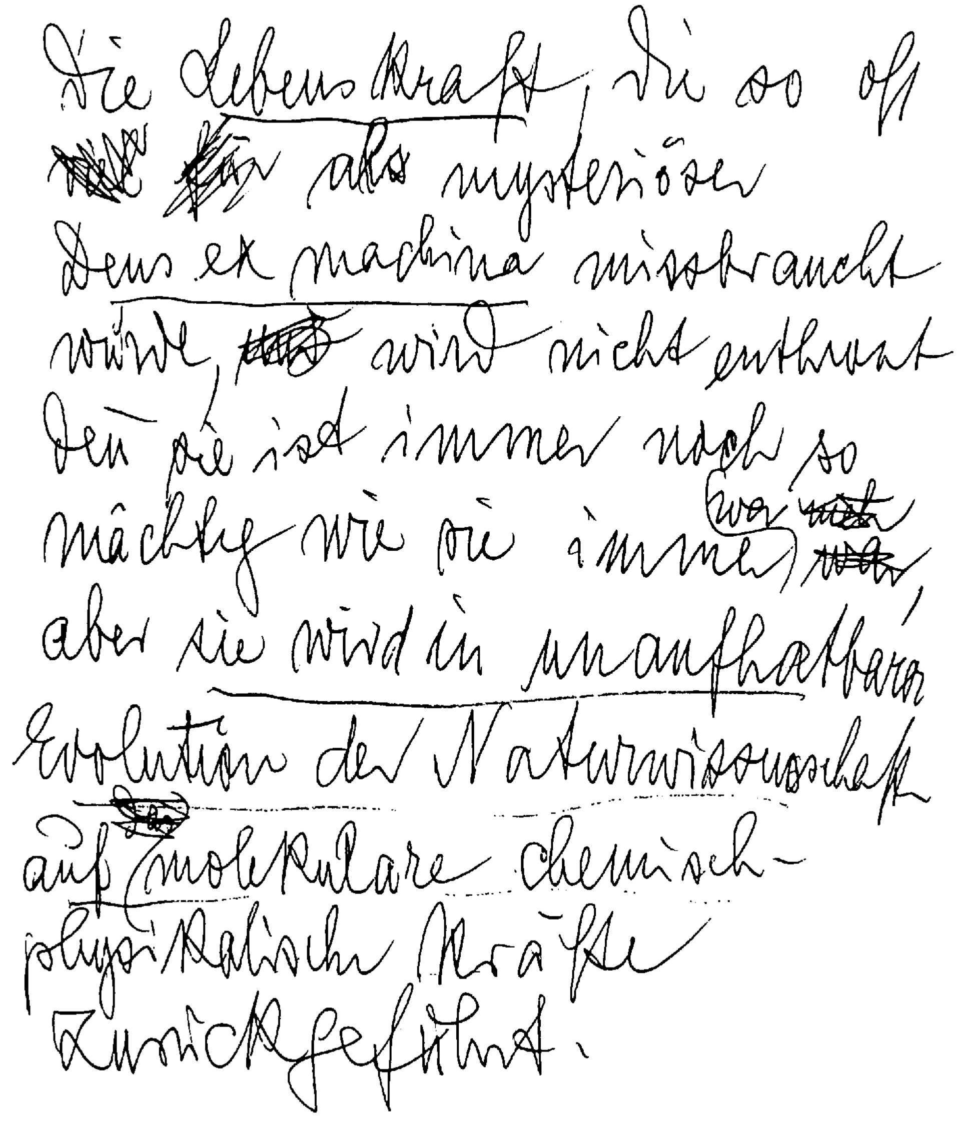
16. Biochemische Evolution

Biochemiker haben seit etwa 30 Jahren begonnen sich ernstlich Gedanken zu machen, nicht nur über den Ursprung des Lebens auf Erden, sondern auch über die Vorgeschichte des Lebens. Oparin und Haldane stellten die plausible Hypothese auf, dass die Vorgeschichte des Lebens in einer biochemischen Evolution bestand. Bevor Leben entstehen konnte, mussten die lebenswichtigen chemischen Substanzen entstanden sein. Die biochemische Evolution muss daher als eine selbstverständliche Voraussetzung der biologischen Evolution angenommen werden. Die biologische Evolution von den niedrigsten Lebewesen aufwärts kann heute kaum mehr bestritten werden. Aber auch ein Gegner der biologischen Evolution wird kaum Einwendungen machen können gegen die biochemische Evolution denn sie besteht ja nur in der Annahme der Entstehung und Evolution organischer Verbindungen ohne Leben. Der heikle Punkt der Gesamt-Evolution besteht darin, dass zwischen der biochemischen und der biologischen Evolution die Entstehung des Lebens liegen muss, ~~gleichgültig ob das Leben "natürlichen" oder "übernatürlichen" Ursprungs ist~~.

CO CO_2
HCl HF
H_2O H_2S
H_3N (H_3P)
H_4C

Für die gesamte Evolution beginnend mit der Entstehung ~~der~~ ersten organischen ~~Moleküle~~ auf Erden steht eine Periode von ungefähr 4000 Millionen Jahren zur Verfügung. Damals bestand die Atmosphäre wohl hautpsächlich aus Methan, Ammoniak CO, CO_2, HCl, HF, H_2S, N_2, NH_4Cl etc und Wasserstoff, auch war viel Waseer vorhanden. Besonders

F In vitro werden beide DNA-Stränge kopiert. Man hat Anhaltspunkte (Ochoa) dass in vivo nur ein DNA-Strang oder nicht komplementäre Portionen beider Stränge kopiert werden.



Die Lebenskraft, die so oft ~~[illegible]~~ ~~für~~ als mysteriöser Deus ex machina missbraucht würde, ~~[illegible]~~ wird nicht entthront, denn sie ist immer noch so mächtig wie sie immer war ~~war~~, aber sie wird in unaufhaltbarer Evolution der Naturwissenschaft auf ~~[illegible]~~ molekulare chemisch-physikalische Kräfte zurückgeführt.

Fig. 28. Reproduction of a loose leaf among the Ruzicka papers.

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[1] V. Prelog, O. Jeger, «Leopold Ruzicka», *Biogr. Mem. Fellows R. Soc. 26* (1980) 411; a German version was published in *Helv. Chim. Acta 66* (1983) 1307.

[2] «Biosynthese heute und während der biologischen und chemischen Evolution», type- and manuscript for a lecture presented at the Meeting of Nobel Laureates, Lindau, July 1967 (Ruzicka Archives, ETH Zürich).

[3] «Biochemie und Weltbild», lecture presented on 18 February 1964 in Bern, organized by the Freistudentenschaft Bern (Ruzicka Archives, ETH Zürich).

[4] L. Ruzicka, «Über das Bauprinzip und die neueste Ausdehnung der Terpenchemie in biologisch wichtige Richtungen», typescript for a lecture presented before the Naturforschende Gesellschaft, Zürich 1933 (Ruzicka Archives, ETH Zürich).

[5] L. Ruzicka, «The Isoprene Rule and the Biogenesis of Terpenic Compounds» with the chapter «Biogenesis of Steroids and Terpenic Compounds» together with A. Eschenmoser and H. Heusser, *Experientia 9* (1953) 357, 362.

[6] A. Eschenmoser, L. Ruzicka, O. Jeger, D. Arigoni, «Eine stereochemische Interpretation der biogenetischen Isoprenregel», *Helv. Chim. Acta 38* (1955) 1890.

[7] a) G. Ohloff, «Der Forscher Leopold Ruzicka als Erfinder», *Chimia 41* (1987) 181, lecture by the president of the Swiss Chemical Society at the Leopold Ruzicka Centennial Symposium, ETH Zürich, 20 March 1987; b) L. Trueb, «Ein Leben für Naturstoffchemie, Alpenblumen und niederländische Malerei – Zum 100. Geburtstag Leopold Ruzickas», *Neue Zürcher Zeitung*, 11 September 1987, p. 25; c) C. Klemm, «Professor Dr. Leopold Ruzicka und seine Stiftung niederländischer Gemälde im Kunsthaus Zürich»; d) H. Gerson, «Leopold Ruzicka als Bildersammler», catalogue of the exhibition «Im Lichte Hollands», Kunstmuseum Basel, 14 June–27 September 1987.

[8] Fragments for a biography entitled: «Vorliebe für die Chemie des Lebens – Leopold Ruzicka im zweiten Halbjahrhundert, 1908–1958, der Strukturchemie», see also the title of one of Ruzicka's Lindau lectures: «Nobelpreise und Chemie des Lebens», Meeting of Nobel Laureates, Lindau, 29 June 1970; see *Naturwiss. Rundsch. 24* (1971) 50. – Cf. facsimile from the manuscript in Fig. 1.

[9] L. Ruzicka, «In the Borderland between Bioorganic Chemistry and Biochemistry», *Ann. Rev. Biochem. 42* (1973) 1.

[10] Cf.[1], p. 441.

[11] Handwritten introduction for a lecture given at a congress in Beograd «Biogenetische Isoprenregel» (approx. 1965).

[12] L. Ruzicka, «Rolle der Riechstoffe in meinem chemischen Lebenswerk», *Helv. Chim. Acta 54* (1971) 1753.

[13] F. Tiemann, P. Krüger, «Über Veilchenaroma», *Ber. Dtsch. Chem. Ges. 26* (1893) 2675, 2691.

[14] L. Ruzicka, exchange of letters with Haarmann und Reimer, chemische Fabrik zu Holzminden G.m.b.H., 1916 (Ruzicka Archives, ETH Zürich).

[15] L. Ruzicka, C.F. Seidel, H. Schinz, «Über die Bruttoformel und einige Umsetzungen des Irons», *Helv. Chim. Acta 16* (1933) 1143.

[16] O. Wallach, «Alicyclic Compounds», in *Nobel Lectures, Chemistry 1901–1921*, Elsevier, Amsterdam (1966), p. 178.

[17] O. Wallach: *Terpene und Campher*, Veit & Co., Leipzig (1909).

[18] F.W. Semmler, *Chem. Ztg. 29* (1905) 1313.

[19] L. Ruzicka, «Die Totalsynthese des Fenchons», *Ber. Dtsch. Chem. Ges. 50* (1917) 1362.

[20] «Über Phenylmethylketen. Von der Grossherzoglich Technischen Hochschule Fridericiana zu Karlsruhe genehmigte Dissertation zur Erlangung der Würde eines Doktor-Ingenieurs, vorgelegt von Leopold Ruzicka, Dipl.-Ing. aus Vukovar (Slavonien)», Karlsruhe 1911.

[21] H. Staudinger, L. Ruzicka, «Insektentötende Stoffe I. Über Isolierung und Konstitution des wirksamen Teils des dalmatinischen Insektenpulvers», *Helv. Chim. Acta 7* (1924) 177.

[22] H. Staudinger, L. Ruzicka, «Insektentötende Stoffe II. Zur Konstitution der Chrysanthemummonocarbonsäure und -dicarbonsäure», *Helv. Chim. Acta 7* (1924) 201.

[23] W.H. Perkin, jun., J.F. Thorpe, *J. Chem. Soc. 75* (1899) 48.

[24] L. Ruzicka, «Perspektiven der Biogenese und der Chemie der Terpene», *Pure Appl. Chem. 6* (1963) 493.

[25] L. Ruzicka, T. Reichstein, R. Pulver, «Synthese des Tetrahydro-artemisiaketons», *Helv. Chim. Acta 19* (1936) 646.

[26] Y. Asahina, S. Tagaki, *J. Pharm. Soc. Japan* (1920) 873.

[27] L. Ruzicka, «History of the Isoprene Rule» (Faraday Lecture), *Proc. Chem. Soc.* (1959) 341.

[28] O. Wallach, «Zur Kenntnis der Terpene und der ätherischen Öle», *Liebigs Ann. Chem. 239* (1887) 1.

[29] L. Ruzicka, «The Life and Work of Otto Wallach» (3rd Pedler Lecture, 10 March 1932), *J. Chem. Soc.* (1932) 1582.

[30] L. Ruzicka, «Zur Kenntnis der Wagnerschen Umlagerung», *Helv. Chim. Acta 1* (1918) 110.

[31] L. Ruzicka, V. Fornasir, «Über die Totalsynthese des Linalools», *Helv. Chim. Acta 2* (1919) 182.

[32] L. Ruzicka, J. Meyer, «Über Sesquiterpenverbindungen I. Überführung des Cadinens in einen Naphthalinkohlenwasserstoff», *Helv. Chim. Acta 4* (1921) 505.

[33] L. Ruzicka, M. Stoll. «Höhere Terpenverbindungen VII. Konstitution des Eudalins,

des Selinens und des α-Santalens. Das Kohlenstoffgerüst der Sesquiterpene», *Helv. Chim. Acta 5* (1922) 923; see also L. Ruzicka, J. Meyer, M. Mingazzini, *ibid. 5* (1922) 345, 353.

[34] R. Robinson, *Annu. Rep. Prog. Chem. 20* (1923) 100.

[35] L. Ruzicka, J. Meyer, «Zur Kenntnis der Abietinsäure. Überführung der Abietinsäure ins Methylreten», *Helv. Chim. Acta 5* (1922) 315, 581.

[36] C. Harries, *Liebigs Ann. Chem. 406* (1914) 173; C. Harries: *Untersuchungen über die natürlichen und künstlichen Kautschukarten,* Springer, Berlin (1919).

[37] H. Staudinger, J. Fritschi, *Helv. Chim. Acta 5* (1922) 785.

[38] L. Ruzicka, G.B.R. de Graaff, H.J. Müller, «Über eine neue Wendung bei der Aufklärung des Kohlenstoffgerüstes der Abietinsäure und der Dextro-pimarsäure», *Helv. Chim. Acta 15* (1932) 1300; L. Ruzicka, H. Waldmann, P.J. Meier, H. Hösli, «Beiträge zur Kenntnis der Lage der Carboxylgruppe und der Doppelbindungen bei der Abietinsäure», *Helv. Chim. Acta 16* (1933) 169.

[39] O. Aschan, *Liebigs Ann. Chem. 424* (1921) 117; A.I. Virtanen, *ibid. 424* (1921) 150.

[40] R.D. Blair, *J. Chem. Soc. (1935)* 1297.

[41] C.K. Ingold, *Annu. Rep. Prog. Chem. 21* (1924) 103.

[42] L. Ruzicka, W. Zimmermann, K. Huber, «Beiträge zur Konstitution von Bestandteilen des Caryophyllengemisches», *Helv. Chim. Acta 19* (1936) 343.

[43] L. Ruzicka, E. Capato, «Ringbildungen bei Sesquiterpenen. Totalsynthese des Bisabolens und eines Hexahydro-cadalins», *Helv. Chim. Acta 8* (1925) 259.

[44] J. Walker, *Chem. Ind.* (1935) 55.

[45] L. Ruzicka, «Die Architektur der Polyterpene», *Angew. Chem. 51* (1938) 5.

[46] L. Ruzicka, «Vielgliedrige Ringe, höhere Terpenverbindungen und männliche Sexualhormone», *Les Prix Nobel en 1945,* Nobel Foundation, Stockholm (1947).

[47] L. Ruzicka, E. Rey, A.C. Muhr, «Über verschiedene Umwandlungsprodukte des Lanosterins», *Helv. Chim. Acta 27* (1944) 472.

[48] W. Voser, M.V. Mijovic, H. Heusser, O. Jeger, L. Ruzicka, «Über die Konstitution des Lanostadienols (Lanosterins) und seine Zugehörigkeit zu den Steroiden», *Helv. Chim. Acta 35* (1952) 2414.

[49] C.S. Barnes, D.H.R. Barton, A.R.H. Cole, J.S. Fawcett, B.R. Thomas, *Chem. Ind.* (1952) 426.

[50] A.R. Penfold, J.L. Simonsen, *J. Chem. Soc.* (1939) 87.

[51] R. Robinson, cited in[50]; see also[53b], p. 12.

[52] K. Bloch, D. Rittenberg, *J. Biol. Chem. 159* (1945) 45; *145* (1942) 625.

[53] a) R. Robinson, «A Theory of the Mechanism of the Phytochemical Synthesis of certain Alkaloids», *J. Chem. Soc.* (1917) 876; b) R. Robinson, «The Structural Relations of Natural Products», First Weizmann Memorial Lectures, December 1953, Clarendon Press, Oxford (1955).

[54] R. Robinson; *Versuch einer Elektronentheorie organisch-chemischer Reaktionen,* Enke, Stuttgart (1932).

[55] F.W. Semmler, H. Stenzel, *Ber. Dtsch. Chem. Ges. 47* (1914) 2555.

[56] L. Ruzicka, J.A. von Melsen, «Höhere Terpenverbindungen XXXIX. Zur Kenntnis des Cedrens», *Liebigs Ann. Chem. 471* (1929) 40.

[57] E. Deussen, *J. Prakt. Chem. [2] 117* (1927) 273.

[58] W. Treibs, *Ber. Dtsch. Chem. Ges. 68* (1935) 1041.

[59] W.F. Short, *Chem. Ind.* (1935) 874.

[60] R. Robinson, J. Walker, *Chem. Ind.* (1935) 906.

[61] L. Ruzicka, E. Jutassy, «Polyterpene und Polyterpenoide CI, Zur Konstitution des Cedrens und Cedrols», *Helv. Chim. Acta 19* (1936) 322.

[62] S.H. Harper, *Annu. Rep. Prog. Chem. 44* (1947) 162.

[63] P.A. Plattner, *Chimia 2* (1948) 248.

[64] A. Eschenmoser, «Zur säurekatalysierten Zyklisierung bei Mono- und Sesquiterpenverbindungen», thesis ETH Zürich No. 2018 (February 1951), Juris-Verlag, Zürich (1952).

[65] P.A. Plattner, A. Fürst, A. Eschenmoser, W. Keller, H. Kläui, S. Meyer, M. Rosner, *Helv. Chim. Acta 36* (1953) 1845.

[66] G. Stork, R. Breslow, *J. Am. Chem. Soc. 75* (1953) 3291.

[67] L. Ruzicka, W. Zimmermann, «Polyterpene und Polyterpenoide XCII. Zur Kenntnis der Caryophyllensäure und der Nor-caryophyllensäure. Über das Additionsprodukt von Maleinsäure-anhydrid an Caryophyllen», *Helv. Chim. Acta 18* (1935) 219.

[68] L. Ruzicka, *Chem. Ind.* (1935) 509.

[69] G.R. Ramage, J.L. Simonsen, *J. Chem. Soc.* (1935) 1581.

[70] H.N. Rydon, *Chem. Ind.* (1938) 123.

[71] F.B. Kipping, *Annu. Rep. Prog. Chem. 35* (1938) 276.

[72] F. Šorm, L. Dolejš, J. Pliva, *Collect. Czech. Chem. Commun. 15* (1950) 186.

[73] D.H.R. Barton, A.S. Lindsay, *Chem. Ind.* (1951) 313.

[74] A. Eschenmoser, H.H. Günthard, *Helv. Chim. Acta 34* (1951) 2338.

[75] T.L. Dawson, G.R. Ramage, B. Wilson, *Chem. Ind.* (1951) 464.

[76] D.H.R. Barton, T. Bruun, A.S. Lindsay, *Chem. Ind.* (1951) 910; *J. Chem. Soc.* (1952) 2210.

[77] F. Zobrist, H. Schinz, *Helv. Chim. Acta 32* (1949) 1192.

[78] P. Dietrich, O. Jeger, «Überführung von Betulin und Oleanolsäure in isomere ungesättigte Kohlenwasserstoffe $C_{29}H_{48}$. Hypothese über die Biosynthese pentacyclischer Triterpene», *Helv. Chim. Acta 33* (1950) 711.

[79] R.G. Langdon, K. Bloch, *J. Biol. Chem. 200* (1953) 129, 135.

[80] M.J.S. Dewar, «The Electronic Theory of Organic Chemistry», Clarendon Press, Oxford (1949).

[81] R.B. Woodward, K. Bloch, *J. Am. Chem. Soc. 75* (1953) 2023.

[82] E.A. Rudolph, «Zur Kenntnis der Azulene und der Gallensäuren», thesis ETH Zürich No. 431 (1925), p. 96–99.

[83] K. Bloch, «The Biological Synthesis of Cholesterol», *Les Prix Nobel en 1964,* p. 179, Nobel Foundation, Stockholm (1965).

[84] R. Robinson, *Chem. Ind.* (1934) 1062.

[85] Personal communication Prof. K. Bloch, Harvard University.

[86] T.T. Tchen, K. Bloch, *J. Am. Chem. Soc. 78* (1956) 1516.

[87] K. Bloch, «Summing up», *Annu. Rev. Biochem. 56* (1987) 1.

[88] E.J. Corey, W.E. Russey, P.R.O. de Montellano, *J. Am. Chem. Soc. 88* (1966) 4750.

[89] E.E. van Tamelen, J.D. Willett, R.B. Clayton, K.E. Lord, *J. Am. Chem. Soc. 88* (1966) 4752.

[90] E. Lederer, D. Mercier, *Experientia 3* (1947) 188.

[91] O. Jeger, O. Dürst, L. Ruzicka, *Helv. Chim. Acta 30* (1947) 1859.

[92] See e.g.[2], p. 11.

[93] J.W. Cornforth, D.G. Hunter, G. Popják, *Biochem. J. 54* (1953) 590, 597.

[94] J.W. Cornforth, *Pure Appl. Chem. 2* (1961) 607.

[95] G. Gamboni, H. Schinz, A. Eschenmoser, «Über den sterischen Verlauf der säurekatalysierten Cyclisation in der Terpenreihe. Cyclisation der *cis*-7-Methyl-octadien-(2,6)-säure-(1)», *Helv. Chim. Acta 37* (1954) 964.

[96] P.A. Stadler, A. Nechvatal, J.A. Frey, A. Eschenmoser, «Untersuchungen über den sterischen Verlauf säurekatalysierter Cyclisationen bei terpenoiden Polyenverbindungen. Cyclisation der 7,11-Dimethyl-2(*trans*),6(*trans*),10-dodecatrien- und der 7,11-Dimethyl-2(*cis*),6(*trans*),10-dodecatriensäure», *Helv. Chim. Acta 40* (1957) 1373.

[97] G. Stork, A.W. Burgstahler, «The Stereochemistry of Polyene Cyclization», *J. Am. Chem. Soc. 77* (1955) 5068.

[98] Lecture notes of Dr. Karl Schenker, postdoctoral fellow with Prof. R.B. Woodward, August 1953–November 1955 (personal communication K. Schenker).

[99] F. Lynen, E. Reichert, L. Rueff, *Liebigs Ann. Chem. 574* (1951) 1.

[100] L.D. Wright, E.L. Cresson, H.R. Skeggs, G.D.E. MacRae, C.H. Hoffman, D.E. Wolf, K. Folkers, *J. Am. Chem. Soc. 78* (1956) 5273.

[101] F. Lynen, H. Eggerer, H. Henning, I. Kessel, *Angew. Chem. 70* (1958) 738.

[102] S. Chaykin, J. Law, A.H. Phillips, T.T. Tchen, K. Bloch, *Proc. Natl. Acad. Sci. USA 44* (1958) 998.

[103] J.W. Cornforth, R.H. Cornforth, G. Popják, I. Youhotsky-Gore, *Biochem. J. 66* (1957) 10P; *ibid. 69* (1958) 146.

[104] R.K. Maudgal, T.T. Tchen, K. Bloch, *J. Am. Chem. Soc. 80* (1958) 2589.

[105] J.W. Cornforth, R.H. Cornforth, A. Pelter, M.G. Horning, G. Popják, *Tetrahedron 5* (1959) 311.

[106] D. Arigoni, «Some Studies in the Biosynthesis of Terpenes and Related Compounds», *Pure Appl. Chem. 17* (1968) 331; «Stereochemical Aspects of Sesquiterpene Biosynthesis», *ibid. 41* (1975) 219.

[107] D.E. Cane, «The Stereochemistry of Allylic Pyrophosphate Metabolism», *Tetrahedron 36* (1980) 1109.

[108] L. Ruzicka, in A. Todd (Ed.): *Perspectives in Organic Chemistry,* Wiley-Interscience, New York (1956), p. 265–314.

[109] L. Ruzicka, fragments of an autobiography (Ruzicka Archives, ETH Zürich).

[110] Letter from the organizers of the lecture to L.R., Freistudentenschaft Bern, 7 February 1964.

[111] J.D. Watson, F.H.C. Crick, «Molecular Structure of Nucleic Acids: A Structure for Deoxyribose Nucleic Acid», *Nature 171* (1953) 737.

[112] S.L. Miller, *Science 117* (1953) 528.

[113] A.I. Oparin, «The Origin of Life», English translation of the original Russian publication of 1924, in J.D. Bernal: *The Origin of Life,* Weidenfeld & Nicholson, London (1967), p. 197.

[114] Proceedings of the First International Symposium on «The Origin of Life on the Earth», Moscow 19–24 August 1957, English-French-German Edition edited for the International Union of Biochemistry, F. Clark, R.L.M. Synge (Ed.), Pergamon Press, Oxford (1959).

[115] M. Calvin, «Die chemische Evolution und der Ursprung des Lebens», *Naturwissenschaften 43* (1956) 387.

[116] M. Calvin, «Chemical Evolution» (The Bakerian Lecture 1965), *Proc. R. Soc. London A 288* (1965) 441; «The Chemistry of Life. How Life Originated on Earth and in the World Beyond», *Chem. Eng. News* (1961) 22 May, 96.

[117] N.H. Horowitz, S.L. Miller, «Current Theories on the Origin of Life», *Fortschr. Chem. Org. Naturst. 20* (1962) 423.

[118] C. Ponnamperuma, «The Chemical Origin of Life», *Sci. J.* (1965) May, 39.

[119] M. Polanyi, «Life transcending Physics and Chemistry», *Chem. Eng. News* (1967) 21 August, 55.

[120] M. Eigen, *Naturwissenschaften 58* (1971) 465.

[121] Cf. «Leopold Ruzicka Centennial Symposium, Zwischenreden–Zwiesprachen», *Chimia 41* (1987) 14.

[122] M. Eigen, P. Schuster, *Naturwissenschaften 64* (1977) 541; *65* (1978) 7, 341.

[123] Cf. e.g. I. Prigogine, I. Stengers, «Order out of Chaos», New Science Library, Shambhala Publ. Inc. Boulder, Colorado (1984).

[124] H. Kuhn, *Naturwissenschaften 63* (1976) 68.

[125] «Evolution», *Nova Acta Leopold. 42,* No. 218, lectures given at the annual meeting, 11–14 October 1973 in Halle (Saale), edited by J.-H. Scharf.

[126] H. Kuhn, in[125], p. 149–163.