109. N.C.Baenziger, Acta Cryst., **4**, 216 (1951).
110. E.R.Lippincott, J.Chem.Phys., **17**, 1351 (1949).
111. D.A.Brown, J.Chem.Phys., **29**, 451 (1958).
112. L.A.Woodward and H.L.Robertz, Trans.Faraday Soc., **52**, 1458 (1956).
113. R.J.Gillespie, J.Chem.Soc., 1002 (1952).
114. R.Ehrlich, A.R.Young, B.M.Lichstein, and D.D.Perry, Inorg.Chem., **2**, 650 (1963).
115. G.Schomburg and E.G.Hoffman, Z.Elektrochem., **61**, 1110 (1957).
116. I.R.Beattie and T.Gilson, J.Chem.Soc., 3528 (1964).
117. E.Bonitz, Ber., **88**, 751 (1955).
118. E.G.Hoffman and G.Schomburg, Z.Elektrochem., **61**, 1101 (1957).
119. H.C.Longuet-Higgins, J.Chem.Soc., 139 (1946).
120. H.W.Schrötter and E.G.Hoffman, Ber., **68**, 627 (1964).
121. H.Ulich and W.Nespital, Z.angew.Chem., **44**, 750 (1931).
122. J.W.Smith, Sci.Progr., **35**, 515 (1947).
123. O.Iamamoto and I.Iamada, Tokyo Kogyo Shikensho Hokoky, **59**, 480 (1964).
124. M.Trefonas and W.N.Lipscomb, J.Amer.Chem.Soc., **81**, 4435 (1959).
125. N.L.Paddock, Nature, **167**, 1070 (1951).
126. G.Jander and K.Kraffczyk, Z.anorg.Chem., **283**, 217 (1956).
127. G.G.Evans, J.K.Kennedy, and F.P.Del Greco, J.Inorg. Nuclear Chem., **4**, 40 (1957).
128. K.Ziegler, R.Köster, H.Lehmkuhl, and K.Reinert, Annalen, **629**, 33 (1960).
129. N.M.Alpatova, O.R.Osipov, and Yu.M.Kessler, Elektrokhimiya, **3**, 102 (1967).
130. S.Glasstone, "Introduction to Electrochemistry" (Translated into Russian), Inostr.Lit., Moscow, 1951.
131. N.L.Paddock, Chem. and Ind., 63 (1953).
132. R.M.Fuoss and F.Accascina, "Electrolytic Conductance", Interscience Publ., New York, 1959.
133. A.M.Sukhtotin, "Voprosy Teorii Rastvorov Elektrolitov v Sredakh s Nizkoi Dielektricheskoi Pronitsaemost'yu" (Problems of the Theory of Solutions of Electrolytes in Media with a Low Dielectric Constant), Goskhimizdat, Leningrad, 1959.
134. N.G.Gaylord, "Reduction with Complex Metal Hydrides" (Translated into Russian), Inostr.Lit., Moscow, 1959.
135. L.W.Trevoy and W.G.Brown, J.Amer.Chem.Soc., **71**, 1675 (1949).
136. H.Nöth, B.P. 820513 (1959); Chem.Abs., **54**, 3885b (1960).
137. O.R.Osipov, N.M.Alpatova, and Yu.M.Kessler, Elektrokhimiya, **2**, 984 (1966).

Institute of Electrochemistry, Academy of Sciences of the USSR

Kurnakov Institute of General and Inorganic Chemistry, Academy of Sciences, Moscow

U.D.C. 547.96

Progress in the Synthesis of Peptides on Polymers

V.S.Vesa

CONTENTS

I. Introduction 114
II. Synthesis of polypeptides on polymers 115
1. Essentials of the method 115
2. Synthesis of polypeptides on polymers in the solid phase 115
3. Synthesis of polypeptides on polymers in the liquid phase 118

I. INTRODUCTION

The synthesis of polypeptides is not only one of the present theoretical problems but also a great scientific task and a great economic undertaking. During the last decade in several countries the synthesis of the less complex polypeptides has become attainable not only in laboratories but also on the industrial scale. These are mainly such important biologically active peptides as the hormones oxytocin and vasopressin, which are now easier to synthesise than to isolate from natural sources. The artificial peptide synthesis was developed so that a few years ago three groups of investigators: Katsoyannis and coworkers[1] (USA), Zahn and coworkers[2] (Federal Rupublic of Germany), and Tsou, Wang, and their coworkers[3] (People's Republic of China) succeeded in synthesising the first simplest protein —the hormone insulin, which consists of 51 amino-acids. The complete synthesis of this hormone was effected in 221 stages. This is indeed a great achievement, which itself characterises the modern level of development of peptide synthesis. However, every synthetic chemist knows very well that the multi-stage nature of a synthesis leads to a sharp drop in the yield of the final compound. Thus it is easy to calculate that in carrying out a twenty-stage synthesis with an 80% yield at each stage, the yield of the final product is only 1.2%. In the example of the synthesis of insulin, where the yields were not 100%, the inadequacy of the existing arsenal of methods of peptide synthesis appeared especially sharply.

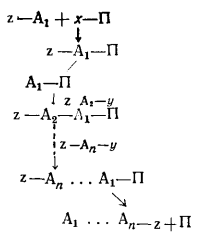
The formation of the peptide bond is only part of the task. A more important obstacle is the purification of the peptide product from impurities formed during the course of the synthesis and which usually have properties very similar to those of the main product. Moreover there is a series of difficulties connected with the low solubility of the peptides in organic solvents, which decreases with increase in the number of amino-acid residues in the molecule. All this placed on the agenda the problem of the creation of more up-to-date methods of synthesising polypeptides. In recent years a way, new in principle, of overcoming the above difficulties has appeared—the construction of peptide chains on three-dimensional polymers in the solid phase and on linear polymers in solution.

II. SYNTHESIS OF POLYPEPTIDES ON POLYMERS

1. Essentials of the Method

In 1959 Merrifield started to develop a new method of synthesising peptides on polymers in the solid phase with the aim of the standardisation and automation of the process. After three years the first report about progress along these lines appeared[4]. Later the synthesis of peptides on polymers was also effected in the liquid phase. The principle of the method is that the first amino-acid $z-A_1$ is attached by a covalent bond to a polymer П. Then amino-acids A_2, A_3, ..., A_n are attached successively to this "anchor" amino-acid. The prepared peptide $z-A_n \ldots A_1-П$ which is formed is separated from the polymer by breaking the peptide–polymer covalent bond at the very end of the synthesis (see Scheme 1, where П is the polymer, z a protective group, A_1, ..., A_n are amino-acids, and x and y are activating groups).

Scheme 1



In the solid-phase synthesis, owing to the insolubility of the peptides attached to the polymer their purification from the starting materials and side-products, even if these impurities are of low solubility, is easily performed by washing with appropriate solvents without danger of loss of peptide. This greatly simplifies the synthesis, shortens the time for carrying out the reaction, and facilitates the purification of the products, so that the standardisation and automation of the processes become possible. The solid phase synthesis allows up to six amino-acid residues to be introduced into a peptide in 24 hours. On using an excess of amino-acid at each stage of the formation of a peptide bond the yield of the peptide usually approaches 100%. Hence the solid-phase synthesis is characterised by simplicity of operation, rapidity, high yields, and the purity of the final products. The synthesis of peptides in the liquid phase on polymers has certain significant differences. Firstly, all the components of the reaction at the time of the synthesis are in the dissolved state but the purification of the polymer–peptide is usually effected by washing free from side-products with water. Secondly, the condensation of amino-acids into peptides is carried out by other methods. In the last stage the peptide separated from the polymer is transferred to an aqueous phase and the polymer remains in an organic layer.

As noted in the very first studies of Merrifield[4,5], the general Scheme 1 is applicable not only to the synthesis of peptides but also to the synthesis of other classes of polymers. Thus, for polynucleotides the individual unit A can be a protected mononucleotide, and for polysaccharides an appropriate protected monosaccharide, etc.

2. Synthesis of Polypeptides on Polymers in the Solid Phase

a. The polymer. In the initial period of the development of the method of peptide synthesis in the solid phase, Merrifield[4,5] carried out considerable work on the selection of a suitable polymer. He tried cellulose, poly-(vinyl alcohol), polymethacrylate, sulphonated polystyrene, and partly chloromethylated copolymer of styrene with 2% divinylbenzene (PSDB–CH_2Cl) which seemed the most suitable for this purpose. Firstly, this polymer is completely insoluble in all the organic solvents used in the synthesis, including neutral, acid, and basic solvents, and at the same time it swells sufficiently in them, which predetermines a rapid penetration of the reagents into the pores of the polymer. Secondly, this polymer has, when swollen, a consistency that is stable and suitable for filtration. Moreover, it contains a functional group capable of reacting with the carboxyl group of the amino-acid or peptide to form an "anchor" covalent bond, which can be split selectively at the end of the peptide synthesis.

b. Protective group and solvent. The success of the synthesis of polypeptides on a polymer basis depends greatly on the choice of a suitable protective group for the amino-acid and of a solvent. In the first studies Merrifield[4,5] protected the amino-components with the benzyloxycarbonyl group (Cbz)†,[6] which is well-known in peptide synthesis and sufficiently readily removable[6]. However, he noted that treatment with hydrogen bromide in glacial acetic acid did not detach all the Cbz groups and that increasing the reaction time led to breaking of some of the "anchor" peptide–polymer ester bonds. For these reasons peptides with shorter chains are formed as impurities and some of the peptide chains are lost. Consequently, it would be necessary either to strengthen the "anchor" benzyl ester bond or to use a more readily removable protective group. Merrifield[4,5] at first followed the first path. For this purpose PSDB–CH_2Cl polymer was nitrated or brominated. As a result of this the loss of peptide due to rupture of the "anchor" benzyl ester bond at the time of removal of the Cbz group became minimal, but the rate of removal of the Cbz groups decreased considerably, especially for the nitrated polymer. On the other hand the polymer–amino-acid bond was strengthened so much that hydrogen bromide in acetic acid had no action on it. Therefore the final peptide was removed from the nitrated polymer by alkaline hydrolysis, and this could lead to a particularly undesirable feature—some racemisation of the peptide. In its properties in peptide syntheses bromination of the polymer occupied an intermediate position.

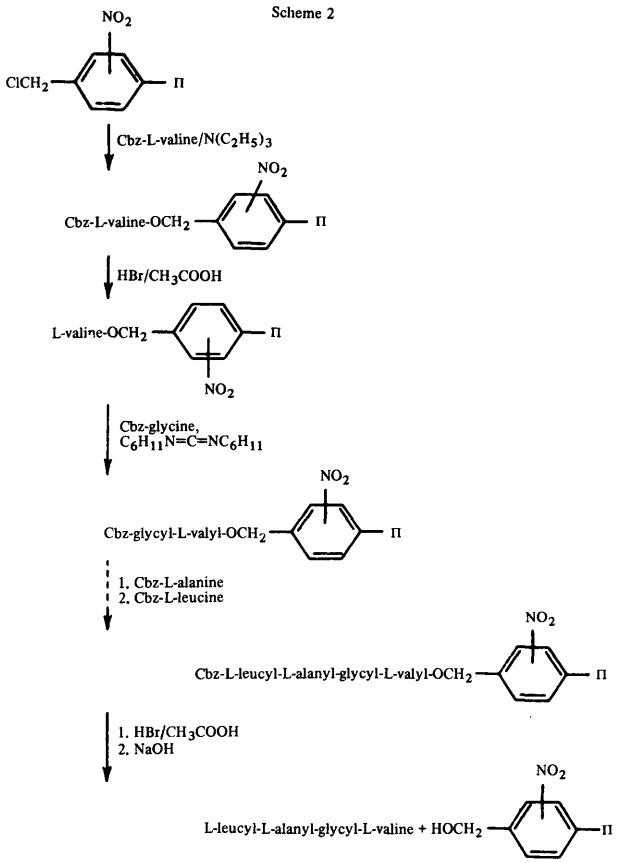
The best solvents in the solid-phase synthesis are dimethylformamide (DMF) and methylene chloride[5], as was shown in studying the synthesis of the tetrapeptide L-leucyl-L-alanyl-glycyl-L-valine on nitrated partially chloromethylated PSDB polymer (Scheme 2, p. 116).

The "anchor" amino-acid Cbz-L-valine was attached by a covalent bond to nitrated PSDB-CH_2Cl polymer by the action of its triethylammonium salt in a suitable solvent; then the protective Cbz group was removed with 10% HBr in acetic acid[7]. After removing all the impurities by washing, DMF was introduced as the best solvent in this synthesis together with Cbz-glycine and the condensing agent, *NN'*-dicyclohexylcarbodi-imide(DCCD). At room

† Cbz is the accepted biochemical abbreviation for the benzyloxycarbonyl (or carbobenzoxy) group (Ed.of Translation).

temperature the reaction lasted 0.5 h. All the side-products, including *NN'*-dicyclohexylurea, were removed by washing with solvents and the Cbz groups were removed as described above. In this cycle the peptide chain is lengthened by one amino-acid. By repeating analogous cycles with Cbz-L-alanine and Cbz-L-leucine the tetrapeptide mentioned above was obtained and was removed from the polymer by hydrolysis with alkali[5], during which partial racemisation of the peptide occurred.

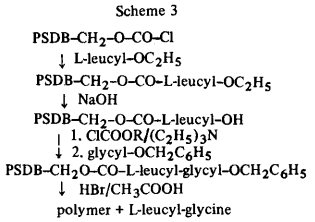
Scheme 2



c. Development of the method. Letsinger and coworkers[8,9] proposed another scheme for the synthesis of peptides in the solid phase. They synthesised a copolymer of styrene with 0.5% divinylbenzene with functional groups which were capable of attachment to the first amino-acid at not the carboxyl, but the amino-group (Scheme 3). This offered the possibility of growing a peptide chain at the unsubstituted carboxyl group of the "anchor" amino-acids by the usual methods. The dipeptide L-leucyl-glycine was obtained in this way.

As we have seen, the strengthening of the "anchor" benzyl ester bond of the polymer–amino-acid did not lead to the desired results. Therefore Merrifield[10] subsequently rejected this path and started to use, instead of a Cbz group, a more readily removable protective group—t-butoxycarbonyl (t-Boc) was introduced into peptide chemistry by McKay and Albertson[11]. In this the solid-phase method not only received a thorough check but was also developed considerably[10,12]. For example, in the synthesis of bradykinin, obtained earlier, particularly by Boissonnas and coworkers[13], polyfunctional amino-acids were included which had been used earlier in solid-phase synthesis: arginine, serine, and proline[10,12]. The solid phase was partly chloromethylated, not nitrated and not brominated, PSDB polymer (Scheme 4). Thanks to the ease of removal of the t-Boc protective groups (1 *N* HCl in acetic acid) the loss of the intermediate peptides was minimised. The use of an excess of t-Boc-amino-acid in the stage of the formation of the peptide allowed the yield of the peptide to be increased and the acetylation with acetic anhydride after each reaction for the formation of the peptide bond previously employed[4,5] to be omitted.

Scheme 3



The final nonapeptide, bradykinin was removed from the solid phase by Gutman and Boissonnas[14] by the action of HBr in trifluoroacetic acid. Note particularly that the synthesis of bradykinin was effected in a special glass apparatus fitted with a system of automatically acting sensors for the initial substances and solvent[15–17]. As a result of the automation of the synthesis the total time required for each cycle was reduced to four hours and the yield of peptide was increased. Thus pure bradykinin with complete biological activity was obtained in 32% yield after 8 days,[10] and in later work[12] in which methylene chloride was used instead of DMF as the solvent, the yield of bradykinin reached 68%.

Subsequently Lenart and coworkers[18,19] proposed to remove the prepared bradykinin from the polymer with dry liquid HF in an excess of anisole. In addition to breaking the peptide–polymer ester bond this also removed the NO_2 groups from arginine.

In seeking antagonists for bradykinin, Stewart and Woolley[20,21] used the automatic synthesis of peptides in the solid phase and within an extremely short time obtained 23 analogues of it, including a bradykinin consisting of D-amino-acids. In a completely analogous manner Merrifield[22] added L-lysine and L-methionine to PSDB–CH_2–O–bradykinin and obtained in 65% yield the chemically pure undecapeptide L-methionyl-L-lysyl-bradykinin, with the complete biological activity of natural kinin. During the synthesis the ϵ-amino-group of lysine was protected with a Cbz, not a t-Boc, group. Alkylation of the sulphide group of methionine with the benzyl bromide formed in the action of HBr in CF_3CO_2H on the Cbz group and *O*-benzylserine was avoided by the presence of ethyl methyl sulphide.

The possibility of including any amino-acid in the peptides was of enormous importance for confirming the general applicability of the new method. With this aim general Scheme 4 was used to synthesise in 56% yield the octapeptide 5-isoleucyl-angiotensin II, which exhibits complete biological activity of the natural material, and twice that of

its analogue[23], and also a hexapeptide[24] in which were successfully included four amino-acids not used earlier in the solid-phase synthesis: asparagine, histidine, isoleucine, and tyrosine. It was shown clearly[24] that if the formation of nitriles is avoided[25] asparagine can be added not by the carbodi-imide method but by Bodanszky's nitrophenyl ester method[26], although this method was rejected by Merrifield after some failures[4,5]. This was confirmed by Bodanszky and Sheehan[27], who found that the nitrophenyl ester method can be used on a par with the carbodi-imide method of amino-acid condensation in solid-phase peptide synthesis. True, they did not give any experimental results in support of this. Merrifield's review[28] of the automatic synthesis of peptides considers not only the carbodi-imide but also Nefkens' hydroxyphthalimide method[29] to be applicable and, exceptionally, in some instances Bodanszky's nitrophenyl ester method[26].

Scheme 4

t-Boc-nitro-L-arginyl-O-CH_2-PSDB

↓ 7 times: 1. 1 *N* HCl/CH_3COOH; 2. $(C_2H_5)_3N$/DMF; 3. t-Boc-amino-acid/DCCD

t-Boc-nitro-L-arginyl-L-prolyl-L-prolylglycyl-L-phenyl-alanyl-*O*-benzyl-L-seryl-L-prolyl-L-phenyl-alanyl-nitro-L-arginyl-O-CH_2-PSDB

↓ HBr/CF_3COOH

Nitro-L-arginyl-L-prolyl-L-prolylglycyl-L-phenylalanyl-L-seryl-L-prolyl-L-phenylalanyl-nitro-L-arginine

↓ H_2, Pd

L-arginyl-L-prolyl-L-prolylglycyl-L-phenylalanyl-L-seryl-L-prolyl-L-phenylalanyl-L-arginine.

Rothe[30] recently published a brief communication stating that he had succeeded in synthesising, on the basis of a Merrifield solid carrier (true, not automatically), a linear oligomer consisting of 14 monomers of ϵ-aminohexanoic acid. Kusch[31,32] gives more detailed information on the synthesis in a solid polymeric phase of a whole series of analogous linear oligomers of hexamethyleneadipic acid amide:

H–[B–A]–OH, where n = 2–5, and 10,

ω-aminoundecanoic acid:

H(Und)–OH, where n = 2–5 and 10,

and ϵ-aminohexanoic acid:

H–(Cap)–OH, where n = 4 and 8,

according to Scheme 5, analogous to the general scheme for synthesis on polymers.

The accumulated experience on the synthesis of peptides on solid carriers allowed Marglin and Merrifield[33] to obtain in sufficiently high yields the A and B chains of insulin in 8 and 11 days respectively. However, completely synthetic insulin exhibited insignificant biological activity, since the A chain contained a racemic asparagine residue.

Considerable difficulties are encountered in including in the peptide chain not only asparagine but also serine and also in the removal of *S*-protected groups in peptides. All these problems which arise in the synthesis of long peptides on carriers were discussed in September 1966 in the Netherlands at the Eighth European Symposium on peptides.

As Katchalski and coworkers showed[34], not only linear but cyclic compounds can be synthesised in the solid phase on polymers. For this purpose poly-(4-hydroxy-3-nitrostyrene) cross-linked with divinylbenzene (I) was obtained, and its active hydroxyl groups were used for preparing high-molecular-weight activated esters of *N*-substituted peptides (II) (Scheme 6).

Scheme 5*

$ClCH_2$– PSDB

↓ t-Boc [B–A]–OH, $(C_2H_5)_3N$

t-Boc –[B–A]–O–CH_2– PSDB

↓ 1. CF_3COOH; 2. $N(C_2H_5)_3$

H–[B–A]–O–CH_2– PSDB

↓ *n* times: 1. t-Boc–[B–A]–OH/DCCD; 2. CF_3COOH; 3. $(C_2H_5)_3N$

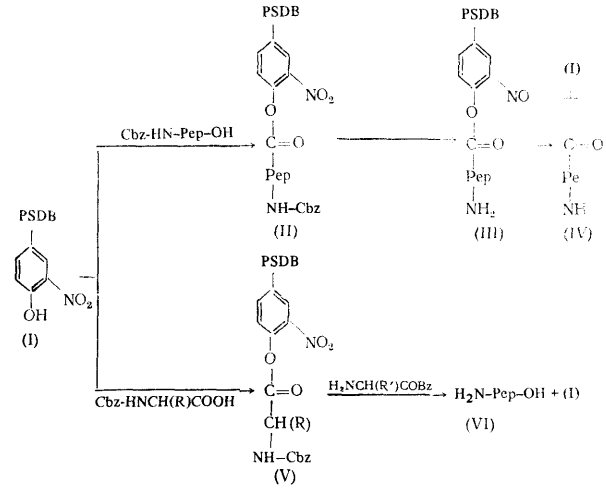
H–[B–A]$_{n+2}$–O–CH_2– PSDB

↓ HBr/CH_3COOH or HBr/CF_3COOH

H–[B–A]$_{n+2}$–OH + $BrCH_2$– PSDB

* –A– = –OC.[CH_2]$_4$.CO–;
–B– = –NH.[CH_2]$_6$NH–;
–Cap– = –HN.[CH_2]$_5$.CO–;
–Und– = –HN.[CH_2]$_5$CO–.

Scheme 6



A high yield of activated ester (II) was obtained only on using at least a triple excess of Cbz–HN–Pep–OH‡ and DCCD. After removing the protective Cbz group the formation of the cyclopeptides (IV) occurred, due to intramolecular aminolysis of the ester bond. This method of cyclisation seemed better than the earlier ones since the final cyclopeptides are obtained pure and in higher yields (60–80%).

Polymer (I) (Scheme 6) can be considered as a chemical reagent-activator which is capable of forming with the carboxyl groups of amino-acids or peptides insoluble activated phenyl ethers (II) and (V). Being activated, the

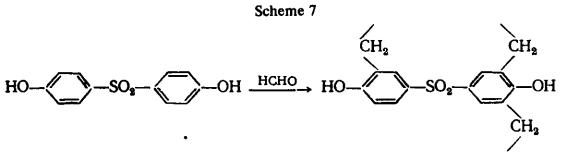
‡ H_2N–Pep–OH is a peptide.

latter can react with compounds which contain unsubstituted amino-groups to form a peptide bond. These considerations were checked experimentally by Katchalski and coworkers [35], and from the insoluble esters (V) Cbz-L-phenylalanine, Cbz-L-proline, etc. were obtained with glycyl-OBz§, S-Cbz-L-cystylglycyl-OBz, etc., the linear peptides (VI) were obtained. At the time of the reaction for the formation of peptides (VI) the initial polymer (I) was liberated and was easily separated from the peptide by centrifuging. By removing the Cbz protective group from the peptide (VI) obtained, it can be introduced again into reaction with the desired insoluble activated polymer with amino-acid (V). On repeating this process a polypeptide of desired structure can be obtained.

Insoluble polymers of phenolic type, one of which contained a mercapto-group in the benzene ring, were made by Shchukina and coworkers [36] and were successfully used for constructing a peptide chain according to Scheme 6.

Wieland and Birr [37] proposed to activate the carboxyl groups of amino-acids with a polymer of phenolic type obtained by the polycondensation of bis-(*p*-hydroxyphenyl) sulphone and formalin according to Scheme 7.

Scheme 7



This insoluble polymer was used to activate Cbz-glycine, t-Boc-L-alanine, etc., which were then used in condensation with the methyl esters of L-phenylalanine and L-valyl-L-phenylalanine.

On detailed examination of all the papers on the synthesis of polypeptides in the solid phase it is easy to note the principal differences in the methods of utilising the polymer phase. In some instances the polymer plays the simultaneous part of a protective group and a peptide carrier from which it is separated at the end of the synthesis, in others the polymer serves as an activating medium and participates in the formation of an amide bond. In the synthesis of peptides by the first path the products were purified by washing the polymer–peptide with solvents and at the end of the synthesis the peptide removed from the polymer was purified in the usual manner. In the second instance the purification of the peptide is possible after any stage, since the peptide which is formed is free from the polymer and present in solution. The synthesis of peptides in the solid phase by the first path, named rightfully after Merrifield, has been greatly standardised and automated. The use of phenol-type polymers for the synthesis of peptides has only started, as we have seen, and will evidently attract even greater attention from investigators in the future.

3. Synthesis of Polypeptides on Polymers in the Liquid Phase

The first studies on the synthesis of peptides on polymers in the solid phase showed certain deficiencies of the method. As we have seen, it can used only when all the reactions are completed rather rapidly and with quantitative yields, since otherwise the final polypeptides separated from the polymer base will be contaminated by peptides of lower molecular weight which are difficult to separate. In turn the rate of formation of peptide bonds and the size of the peptide depends on the permeability of the polymer by amino-acids, peptides, condensing agents, etc. This property is very specific for every concrete instance taken and depends on a series of factors, often not predictable.

With the aim of surmounting the above drawbacks of the solid-phase method, Shemyakin and coworkers [38] proposed the synthesis of polypeptides on linear polymers in the liquid phase. Here the polymer and the growing peptide chain attached to it, just as all the agents, are found in the dissolved state. Partially chloromethylated polystyrene was chosen as the polymeric base (scheme of synthesis analogous to Scheme 2). t-Boc-glycine was attached to it by an ester bond. Then a peptide chain was constructed at room temperature by the method of hydroxysuccinimide esters of t-Boc-amino-acids [39] in DMF. The removal of the excess of the agents and impurities was carried out by washing the precipitated polymer with water and the final tetrapeptide glycyl-glycyl-L-leucyl-glycine, obtained in 65% yield, was detached from the polymer by HBr in CF_3CO_2H.

A second example of the synthesis of peptides, but already cyclic peptides, by the liquid-phase method is the work of Katchalski and coworkers [34]. They synthesised the soluble branched copolymer of L-lysine with 3-nitro-L-tyrosine in which the free amino-groups were blocked by acetylation, and the hydroxyl groups remained free, capable of forming ester bonds with *N*-substituted amino-acids or peptides. However, this polymer had one significant drawback since the high yields of Cbz-*N*-activated esters of type (II) (Scheme 6) could be obtained by using only a considerable excess of Cbz-*N*-peptide. In this connection additional difficulties were created, connected with the removal of the excess of unreacted peptides from a solution. After removing the protective group and the neutralisation of the corresponding hydrobromides an internal aminolysis took place and cyclopeptides were formed in 60–80% yield. In this way cyclo-(glycyl-glycyl), cyclo-(tetra-L-alanyl), etc. were obtained. However, these contained peptide oligomers as impurities, which were not observed in the solid-phase synthesis of cyclopeptides [34].

---o0o---

In the development of the new method of the synthesis of peptides on polymers in the solid phase the first substantial successes have been obtained. This relates especially to syntheses on PSDB–CH_2Cl polymer, with the use of which the conditions for the combination into peptides of many amino-acids have been studied, including polyfunctional amino-acids, and several linear oligomers of the nylon type have been obtained. It is remarkable that already bradykinin, methionyl-lysyl-bradykinin, angiotensin II, 5-isoleucyl-angiotensin, and several analogues of bradykinin of the L- and D-series have been synthesised. The complete biological activity of the above synthesised products of the L-series confirms the extremely important fact that racemisation does not take place during their synthesis in the solid phase. The high rate and the economic nature of the synthesis results from its considerable standardisation and automation. However, as already noted, the method also has some significant drawbacks and limitations concerning the inclusion in the peptides of asparagine, serine, the removal of *S*-protective groups,

§ Bz = benzyl.

etc. All these problems are now being solved by investigators.

As Merrifield noted[4,5], the solid-phase method allows not only peptides to be constructed on a polymer, but also other oligomers, for example, nucleotides, the first papers about the synthesis of which have already appeared[40-43]. All this suggests that the solid-phase method has enormous prospects. It helps investigators to obtain compounds of the most varied degrees of complexity, having inestimable value not only for theory but also for practice.

Owing to the insufficient information it is at present difficult to state definitely the advantages of the liquid-phase method in comparison with the solid-phase. It must be noted that good results were obtained with this method in the synthesis of oligonucleotides[44,45]. Undoubtedly further investigations in this direction will bring new valuable results.

REFERENCES

1. P.G.Katsoyannis, F.Fukuda, A.Tometsko, K.Suzuki, and M.Tilak, J.Amer.Chem.Soc., **86**, 930 (1964).
2. J.Meienhofer, E.Schnabel, H.Bremer, O.Brinkhoff, R.Zabel, W.Stroka, H.Klostermeyer, D.Brandenburg, T.Okuda, and H.Zahn, Naturforsch., **18b**, 1130 (1963).
3. C.J.Niu, Y.T.Kung, W.T.Huang, L.T.Ke, C.C.Chen, Y.C.Chen, Y.C.Du, R.Q.Jiang, C.L.Tsou, S.C.Hu, S.Q.Chu, and K.Z.Wang, Sci.Sinica (Peking), **13**, 1343 (1964).
4. R.B.Merrifield, Fed.Proc., **21**, 412 (1962).
5. R.B.Merrifield, J.Amer.Chem.Soc., **85**, 2149 (1963).
6. M.Bergmann and L.Zervas, Chem.Ber., **65**. 1192 (1932).
7. D.Ben-Ishai and A.Berger, J.Org.Chem., **17**, 1564 (1952).
8. R.L.Letsinger and M.J.Kornet, J.Amer.Chem.Soc., **85**, 3045 (1963).
9. R.L.Letsinger, M.J.Kornet, V.Mahadevan, and D.M.Jerina, J.Amer.Chem.Soc., **86**, 5163 (1964).
10. R.B.Merrifield, J.Amer.Chem.Soc., **86**, 304 (1964).
11. F.C.McKay and N.F.Albertson, J.Amer.Chem.Soc.. **79**, 4686 (1957).
12. R.B.Merrifield, Biochemistry, **3**, 1385 (1964).
13. R.A.Boissonnas, S.Gutmann, and P.A.Jacquenoud, Helv. Chim.Acta, **43**, 1349 (1960).
14. S.Gutmann and R.A.Boissonnas Helv.Chim.Acta, **42**, 1257 (1959).
15. R.B.Merrifield and J.M.Stewart, Nature, **207**, 522 (1965).
16. R.B.Merrifield and J.M.Stewart, Chem.Eng.News, **43** (25), 40 (1965).
17. C.Male, New Scientist, **27**, 337 (1965).
18. J.Lenart and A.B.Robinson, J.Amer.Chem.Soc., **89**, 181 (1967).
19. J.Lenart, J.Org.Chem., **32**, 250 (1967).
20. J.M.Stewart and D.W.Woolley, Nature, **206**, 619 (1965).
21. J.M.Stewart and D.W.Woolley, Fed.Proc., **24**, 657 (1965).
22. R.B.Merrifield, J.Org.Chem., **29**, 3100 (1964).
23. G.R.Marshall and R.B.Merrifield, Biochemistry, **4**, 2394 (1965).
24. D.W.Woolley, J.Amer.Chem.Soc., **88**, 2309 (1966).
25. D.T.Gish, P.G.Katsoyannis, G.P.Hess, and R.J.Stedman, J.Amer.Chem.Soc., **78**, 5954 (1956).
26. M.Bodanszky, Nature, **175**, 685 (1955).
27. M.Bodanszky and T.T.Sheehan, Chem.and Ind., 1423 (1964).
28. R.B.Merrifield, Science, **150**, 178 (1965).
29. G.H.I.Nefkens and G.I.Tesser, J.Amer.Chem.Soc.. **83**. 1263 (1961).
30. M.Rothe, Angew.Chem., **78**, 151 (1966).
31. P.Kusch, Kolloid-Z., **208** (2), 138 (1966).
32. P.Kusch, Angew.Chem., **78**, 611 (1966).
33. A.Marglin and R.B.Merrifield, J.Amer.Chem.Soc., **88**, 5051 (1966).
34. M.Fridkin, A.Patchornik, and E.Katchalski, J.Amer. Chem.Soc., **87**, 4646 (1965).
35. M.Fridkin, A.Patchornik, and E.Katchalski, J.Amer. Chem.Soc., 88, 3164 (1966).
36. L.Yu.Sklyarov, V.I.Gorbunov, and L.A.Shchukina, Zhur. Obshch.Khim., **36**, 2220 (1966).
37. T.Wieland and C.Birr, Angew.Chem., **78**, 303 (1966).
38. M.M.Shemyakin, Y.A.Ovchinikov, A.A.Kiryuchkin, and I.V.Kozhevnikova, Tetrahedron Letters, **27**, 2323 (1965).
39. G.W.Anderson, J.E.Zimmerman, and F.M.Callahan, J.Amer.Chem.Soc., **86**, 1839 (1964).
40. R.L.Letsinger, J.Fontaine, V.Mahadevan, D.A.Schexnayder, and R.E.Leone, J.Org.Chem., **29**, 2615 (1964).
41. R.L.Letsinger and V.Mahedevan, J.Amer.Chem.Soc., **87**, 3526 (1965).
42. R.L.Melby and D.R.Strobach, J.Amer.Chem.Soc., **89**, 450 (1967).
43. G.M.Blackburn, M.J.Brown, and M.R.Harris, Chem. Comm., No.17, 661 (1966).
44. H.Hayatsu and H.G.Khorana, J.Amer.Chem.Soc., 88, 3182 (1966).
45. F.Cramer, Angew.Chem., **78**, 640 (1966).

Institute of Chemistry and Chemical Technology, Academy of Sciences of the Lithuanian SSR, Vilna

---

U.D.C. 547.31

The Structure of Conjugated Open-chain Hydrocarbons

E.M.Popov and G.A.Kogan

CONTENTS

I. Introduction	119
II. Basic concepts of the structure of organic compounds	120
III. Quantum-mechanical studies of conjugated compounds	124
IV. The vibrational spectra of conjugated compounds	129

I. INTRODUCTION

One of the most important problems in theoretical organic chemistry is the study of the electron density distribution in molecules. Systems with conjugated carbon–carbon bonds occupy a unique position in this problem. Modern theoretical methods of organic chemistry have been their greatest successes when applied to such molecules as polyenes, poly-ynes, aromatic systems (benzenoid and non-benzenoid), organic dyes, and cumulenes. The