453. *Purines, Pyrimidines and Glyoxalines. Part V.* New Syntheses of Uracils and Orotic Acids.*

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Syntheses of a number of substituted uracils and orotic acids from *N*-ethoxycarbonyl derivatives of propiolamide, β-ethoxyacrylamide, and maleic anhydride have been developed. A method of decarboxylating orotic acid and its derivatives is described.

Earlier papers in this series have been partly concerned with the synthesis of 5-cyanouracils by reaction of a primary amine with the ethoxy-derivative (I; R = CN). Replacement of the cyano-group in these pyrimidines by hydrogen may be effected by acid hydrolysis and decarboxylation of the resulting 5-carboxylic acid. Two examples are described in the Experimental section. Such methods, however, would not be applicable to cyanouracils bearing acid-labile substituents, in particular sugar residues, and, in an attempt to widen the scope of our pyrimidine syntheses, we considered alternative routes to 1-substituted uracils from simple precursors, possibly analogous to those which might conceivably arise in biochemical systems. We now report some preliminary experiments

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leading to the synthesis of simple precursors of uracil and uracil-6-carboxylic acid (orotic acid) derivatives.

Propiolic anhydride reacted remarkably slowly with urethane under a variety of conditions, to give eventually the propiolamide (II) which was accompanied by two isomeric substances formed by addition of urethane to the amide (II), and presumably *cis-trans*-isomers or α- and β-adducts. The propiolamide reacted vigorously with aniline at room temperature, to give the linear anilinoacrylamide (III; R = Ph) which with dilute alkali readily afforded 1-phenyluracil (V; R = Ph), also obtained from 5-cyano-1-phenyluracil by hydrolysis and decarboxylation.

(I) $R{\cdot}C(:CH{\cdot}OEt){\cdot}CO{\cdot}NH{\cdot}CO_2Et$ $HC{\vdots}C{\cdot}CO{\cdot}NH{\cdot}CO_2Et$ (II)

(III) $R{\cdot}NH{\cdot}CH{:}CH{\cdot}CO{\cdot}NH{\cdot}CO_2Et$ $EtO{\cdot}CH{:}CH{\cdot}CO_2Et$ (IV)

Reaction of ethyl propiolate with the sodium derivative of urethane gave a readily separable mixture of the β-ethoxyacrylamide (I; R = H) and ethyl β-ethoxyacrylate (IV). The ethoxyacrylamide (I; R = H) with ammonia, methylamine, and aniline followed by dilute alkali readily gave uracil, 1-methyluracil, and 1-phenyluracil (V; R = H, Me, and Ph) respectively.

Synthesis of orotic acid (VI; R = H) and derivatives by our present methods required an oxaloacetylurethane or its equivalent. Oxaloacetic acid and urethane in the presence of phosphoryl chloride or acetic anhydride gave a good yield of *N*-ethoxycarbonylaminomaleic anhydride (VII), whose structure is confirmed by hydrogenation followed by hydrolysis to aspartic acid, and by its further reactions. The anhydride with ammonia gave almost quantitatively a compound, presumably the maleimide (VIII; R = H), which with dilute alkali gave 5-carboxymethylenehydantoin (IX; R = R′ = H); this compound was converted into orotic acid when warmed with alkali (cf. Mitchell and Nyc [1]). Similarly the anhydride with aniline and sulphanilamide gave maleimides (VIII; R = Ph and *p*-$H_2N{\cdot}SO_2{\cdot}C_6H_4$) which when heated with alkali gave the 3-substituted orotic acids (VI; R = Ph or *p*-$H_2N{\cdot}SO_2{\cdot}C_6H_4$). A phenylorotic acid, identical with our material, has been prepared by the reaction of phenylurea with diethyl oxaloacetate and hydrolysis; [2] the intermediate product, assumed to be the ethyl ester of phenylorotic acid, is undoubtedly the hydantoin ester (IX; R = Ph, R′ = Et) since Mitchell and Nyc [1] have shown that condensation of urea with diethyl oxaloacetate gave the hydantoin ester (IX; R = H, R′ = Et). The position of the phenyl group in these compounds was based on experiments by Bachstez who prepared several alkylorotic acids

Ultraviolet absorptions.

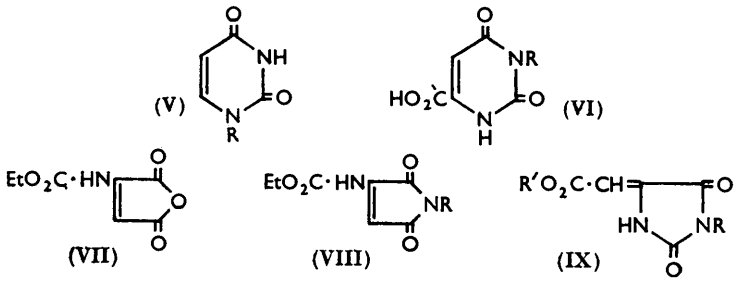
	(IX; R′ = Et)		(IX; R′ = H)		(VI)	
R	$\lambda_{max.}$ (mμ)	$10^{-3}\varepsilon$	$\lambda_{max.}$ (mμ)	$10^{-3}\varepsilon$	$\lambda_{max.}$ (mμ)	$10^{-3}\varepsilon$
H	300	13	290	11·8	278	6·4
Ph	278	12·6	282	15·2	280	6·7
Me	300	9·65	—	—	280	7·4

by the similar reaction of an alkylurea with diethyl oxaloacetate,[3] and the position of the substituent was assigned only by analogy with the reaction of alkylureas with ethyl acetoacetate. An alternative structure for the above-mentioned phenylorotic acid would be the carboxyhydantoin (IX; R = Ph, R′ = H). This compound has now been prepared from oxaloacetic acid and phenylurea and shown to differ from the phenylorotic prepared as above. In addition, the light absorption (see Table) of some orotic derivatives and analogous hydantoins clearly differentiates the two series, the hydantoins showing enhanced absorption.

Final confirmation of the structures of both our phenyl orotic acid and the methylorotic acid prepared by Bachstez's method came by decarboxylation of the acids in

[1] Mitchell and Nyc, *J. Amer. Chem. Soc.*, 1947, **69**, 674.
[2] Ridi and Aldo, *Gazzetta*, 1952, **82**, 23.
[3] Bachstez, *Ber.*, 1931, **64**, 2683.

quinoline in the presence of copper powder to the known 3-phenyl- and 3-methyl-uracil. Orotic acid was also decarboxylated under these conditions to uracil. These appear to be the first recorded successful decarboxylations of orotic acid and its derivatives *in vitro*



although the biochemical decarboxylation of orotic acid is well known and of considerable importance.

Experimental

Unless otherwise indicated, compounds were dried for analysis at 55°/0·1—0·5 mm. over phosphoric oxide.

Reaction of Propiolic Anhydride with Urethane.—Propiolic anhydride[4] (7 g.) and urethane (5·1 g.) were heated together at 95° (internal temp.) for 2 hr. The product, which still contained excess of anhydride, was extracted with ether (100 ml.), and the extract washed with water (50 ml.) and 5% aqueous sodium hydrogen carbonate (2 × 50 ml.). The combined aqueous solutions were extracted with ether (50 ml.). From the dried ether solutions by evaporation was obtained a semicrystalline syrup which when treated with hot benzene left a *substance* (0·26 g.) which separated from chloroform as needles, m. p. 188° (Found: C, 46·8; H, 6·3; N, 12·25. $C_5H_{12}O_5N_2$ requires C, 46·95; H, 6·1; N, 12·2%). An *isomer* (0·7 g.) separated from the cooled benzene solution as prisms, m. p. 145° (Found: C, 47·1; H, 6·3; N, 12·3%). The benzene solution was finally evaporated and the residue distilled *in vacuo*, to give N-*ethoxycarbonylpropiolamide* (1·8 g.), b. p. 108—114°/4 mm., which crystallised, and recrystallised from benzene–light petroleum as laths, m. p. 74° (Found: C, 51·1; H, 5·05; N, 10·15. $C_6H_7O_3N$ requires C, 51·1; H, 5·0; N, 9·9%).

Reaction of Ethyl Propiolate with Sodium Urethane.—Urethane (5 g.) in ether (150 ml.) was heated under reflux with sodium (1·3 g.) for 5 hr. Ethyl propiolate (5·5 g.) in benzene (50 ml.) was added and the suspension was stirred and boiled under reflux for 60 hr. The resulting dark-brown solution was washed with ice-cold N-sulphuric acid (150 ml.); the aqueous phase was extracted with ether, and the combined ether solutions were dried and evaporated to an oil which was distilled *in vacuo*, to give fractions b. p. 40—80°/1 mm. and 120—126°/1 mm. The first fraction was redistilled and ethyl β-ethoxyacrylate (1 g.), b. p. 194°, was collected (Found: C, 58·1; H, 8·6. Calc. for $C_7H_{12}O_3$: C, 58·3; H, 8·4%). Deno[5] records b. p. 189—193°. The second fraction crystallised and after sublimation at 80—100°/0·1 mm. gave β-*ethoxy*-N-*ethoxycarbonylacrylamide* (1·6 g.), m. p. 84° (Found: C, 51·6; H, 6·9; N, 7·6. $C_8H_{13}O_4N$ requires C, 51·3; H, 7·0; N, 7·5%).

1-*Phenyluracil.*—(*a*) When *N*-ethoxycarbonylpropiolamide (0·12 g.) and aniline (0·08 g.) were mixed a vigorous reaction occurred. The cooled product crystallised and was triturated with ether, to give β-*anilino*-N-*ethoxycarbonylacrylamide* (0·16 g.) which separated from light petroleum (b. p. 60—80°) as cream-coloured prisms, m. p. 146—148° (Found: C, 61·1; H, 5·7; N, 12·0. $C_{12}H_{14}O_3N_2$ requires C, 61·5; H, 6·0; N, 12·0%). *N*-Ethoxycarbonylpropiolamide (0·16 g.) and aniline (0·11 g.) were treated as in the previous experiment; the product dissolved in 2N-potassium hydroxide (3 ml.) when shaken for a few min. Acidification gave a solid precipitate; 1-*phenyluracil* (0·1 g.) separated from water as needles, m. p. 246° (Found: C, 63·7; H, 4·1; N, 14·75. $C_{10}H_8O_2N_2$ requires C, 63·8; H, 4·3; N, 14·9%). In

[4] Straus and Voss, *Ber.*, 1926, **59**, 1681.
[5] Deno, *J. Amer. Chem. Soc.*, 1947, **69**, 2233.

admixture with 3-phenyluracil,[6] m. p. 244°, the compound had m. p. 210—220°. (*b*) 5-Cyano-1-phenyluracil [7] (2 g.) was boiled under reflux with 6N-hydrochloric acid (200 ml.) for 12 hr. The solution was concentrated to 50 ml., filtered hot, and cooled, a solid precipitate being obtained. This was washed with water, then kept with saturated aqueous sodium hydrogen carbonate (60 ml.) for 3 days. An insoluble sodium salt remained which was dissolved in water, and the solution acidified to give *1-phenyluracil-5-carboxylic acid* (0·4 g.) which separated from water as needles, m. p. 274° (decomp.) (Found: C, 57·2; H, 3·7; N, 12·6. $C_{11}H_8O_4N_2$ requires C, 56·9; H, 3·5; N, 12·1%). A sample was heated at the m. p. until effervescence ceased. The residue, crystallised from water, had m. p. 240° alone or mixed with 1-phenyluracil. Acidification of the sodium hydrogen carbonate solution precipitated unchanged 5-cyano-1-phenyluracil (0·2 g.), m. p. and mixed m. p. 280—283° (Found: C, 61·7; H, 3·6; N, 19·4. Calc. for $C_{11}H_7O_2N_3$: C, 61·95; H, 3·3; N, 19·7%). (*c*) β-Ethoxy-*N*-ethoxycarbonylacrylamide (0·05 g.) was gently warmed for 5 min. with aniline (0·35 ml.). The product was shaken with N-sodium hydroxide (1 ml.) on a water-bath for 5 min., and the excess of aniline removed by extraction with ether. Acidification of the aqueous solution gave a precipitate of 1-phenyluracil (0·05 g.), m. p. and mixed m. p. 244—245°. (*d*) Urethane (1·2 g.) was converted into the sodium derivative as in the above experiment. To the ethereal suspension was added ethyl propiolate (1·3 g.), and the mixture was boiled under reflux for 18 hr., cooled, washed with cold hydrochloric acid, dried, and evaporated to an oil. This was treated with aniline (1·25 ml.) and kept at 90° for 1 hr., and the resulting crystalline product shaken with 2N-sodium hydroxide (10 ml.) for 10 min. Excess of aniline was removed by extraction with ether, and the aqueous solution acidified with hydrochloric acid to give 1-phenyluracil (0·9 g.), m. p. and mixed m. p. 244°. Smaller yields of the same material were obtained when an excess of urethane was used or when the reactants were shaken together at room temperature for 16 hr. In the latter case a cleaner product was obtained initially.

Uracil and 1-Methyluracil.—(*a*) An ethereal solution of β-ethoxy-*N*-ethoxycarbonylacrylamide was prepared as under (*d*) above from urethane (2 g.). This was treated with 2N-aqueous ammonia (25 ml.), and the mixture evaporated to dryness at 60°. The residue was mixed with dilute sodium hydroxide solution and, after extraction with ether, acidified to give a precipitate of uracil (0·1 g.), m. p. and mixed m. p. 315—320°. (*b*) The intermediate prepared as in (*a*) from urethane (4 g.) was warmed with 33% ethanolic methylamine (5 ml.) at 60° for 20 min., then evaporated to dryness. The residue was warmed for 5 min. with 2N-sodium hydroxide (5 ml.), extracted with ether, and acidified. The solution was evaporated to dryness *in vacuo* and the residue extracted with *iso*butyl ketone (3 × 5 ml.); 1-methyluracil (0·2 g.) crystallised from the extract, having m. p. and mixed m. p. 233° (Found: C, 47·6; H, 4·8; N, 22·2; Calc. for $C_5H_6O_2N_2$: C, 47·6; H, 4·8; N, 22·2%). (*c*) 5-Cyano-1-methyluracil [7] (2·5 g.) was boiled under reflux with 6N-hydrochloric acid (15 ml.) for 8 hr. The solution was evaporated to dryness and the residue dissolved in aqueous sodium hydrogen carbonate. The acidified solution was extracted with ethyl acetate and from the organic phase was recovered *1-methyluracil-5-carboxylic acid* (1·8 g.) which recrystallised from water as prisms, m. p. 266° (decomp.) (Found: C, 42·1; H, 3·6; N, 16·5. $C_6H_6O_4N_2$ requires C, 42·4; H, 3·6; N, 16·5%). A small quantity of the acid was heated at 260—280° for 10 min.: it effervesced and a sublimate was obtained. This was resublimed at 150°/0·4 mm. to give 1-methyluracil, m. p. and mixed m. p. 231—232° (Found: N, 22·4%); Brown, Hoerger, and Mason [8] record m. p. 232—233°.

Reaction of Oxaloacetic Acid with Urethane.—Urethane (1·8 g.) and oxaloacetic acid (2·6 g.) in phosphoryl chloride (6 ml.) were heated at 70° until a clear solution was obtained (30 min.). This was cooled to give a crystalline precipitate which was filtered off and washed with ice-water (10 ml.). *N-Ethoxycarbonylaminomaleic anhydride* (2 g.) separated from ethyl acetate–light petroleum as needles, m. p. 118° (Found: C, 45·7; H, 4·1; N, 7·6. $C_7H_7O_5N$ requires C, 45·4; H, 3·8; N, 7·6%). The same compound was also obtained in lower yield by the reaction between urethane, oxaloacetic acid, and acetic anhydride. The anhydride (0·18 g.) in ethanol (20 ml.) was hydrogenated over platinum oxide for 3 hr., hydrogen absorption then ceasing. The solution was filtered and evaporated to dryness. The residue was boiled under reflux with N-hydrochloric acid (10 ml.) for 1 hr. Paper chromatograms of the solution were run, solvent being the upper layer of a mixture of butanol, water, and acetic acid (25 : 25 : 6).

[6] Whitehead, *J. Amer. Chem. Soc.*, 1952, **74**, 4267.
[7] Shaw, *J.*, 1955, 1834.
[8] Brown, Hoerger, and Mason, *J.*, 1955, 211.

A strong ninhydrin-positive spot was obtained indistinguishable from that of aspartic acid run under identical conditions.

Orotic Acid.—The foregoing anhydride (0·3 g.) was dissolved in saturated ethanolic ammonia (5 ml.). After the initial spontaneous evolution of heat had ceased the solution was cooled to 0°; N-*ethoxycarbonylaminomaleimide* (0·28 g.) crystallised (m. p. 85°) (Found: N, 15·3. $C_7H_8O_4N_2$ requires N, 15·2%). The compound was unstable and deteriorated during several days at room temperature. The maleimide (0·51 g.) was dissolved in 2N-aqueous sodium hydroxide (3 ml.); the solution was warmed for 3 min. at 50°, cooled, and acidified to precipitate 5-carboxymethylenehydantoin (0·3 g.) which recrystallised from water as plates, decomp. from 300° (Found: C, 38·6; H, 2·7; N, 18·2. Calc. for $C_5H_4O_4N_2$: C, 38·5; H, 2·6; N, 17·95%); Mitchell and Nyc [1] record m. p. 300—400° (decomp.). As recorded by the latter authors, the hydantoin was readily converted into orotic acid. The hydantoin (10 mg.) was heated for 30 min. with N-potassium hydroxide (0·25 ml.), and the cooled solution acidified with hydrochloric acid to precipitate orotic acid monohydrate (9 mg.), m. p. 336° undepressed when admixed with a sample prepared from 6-methyluracil by Behrend and Struve's method.[9] Both compounds also moved as single spots (R_F 0·46) in propan-1-ol, formic acid, water (7 : 1 : 2).

3-*Phenylorotic Acid.*—(*a*) Aniline (0·1 g.) was added to a solution of the anhydride (0·17 g.) in ethanol (1 ml.). The solution was warmed for a few min. to give a yellow solution which when cooled gave a crystalline precipitate. α-N-*Ethoxycarbonylamino*-N′-*phenylmaleimide* (0·1 g.) crystallised from ethanol as needles, m. p. 135° (Found: C, 59·95; H, 4·95; N, 11·1. $C_{13}H_{12}O_4N_2$ requires C, 60·0; H, 4·65; N, 10·8%). Aqueous sodium hydroxide was added gradually to a suspension of the phenylmaleimide (1 g.) in water (3 ml.) to give a clear solution. This was cooled and acidified with 10N-hydrochloric acid, giving a crystalline precipitate; 3-phenylorotic acid monohydrate (0·64 g.) recrystallised from ethanol as needles, m. p. 280° (decomp. with loss of solvent from 180°) (Found: C, 53·1; H, 4·1; N, 11·15. Calc. for $C_{11}H_8O_4N_2,H_2O$: C, 52·8; H, 4·0; N, 11·2%). The compound was recovered unchanged after treatment with N-potassium hydroxide at 100° for 30 min. (*b*) The anhydride (0·5 g.) and aniline (3 ml.) were heated together for 10 min. at 60° and the product stirred with 2N-sodium hydroxide (2·5 ml.) for a further 10 min. Unchanged aniline was removed by extraction with ether and 3-phenylorotic acid monohydrate (0·31 g.) was recovered by acidification of the alkaline solution. The acid had m. p. and mixed m. p. 280° (decomp.) (Found: C, 52·9; H, 4·0; N, 11·2%). (*c*) To a solution of 5-carbethoxymethylene-3-phenylhydantoin (prepared from diethyl oxaloacetate and phenylurea) (1·44 g.) in boiling ethanol (100 ml.) was added a solution of potassium hydroxide (0·6 g.) in water (3 ml.). The yellow solution was cooled and treated with water (34 ml.), then evaporated *in vacuo* to a small volume. This, when acidified with hydrochloric acid, precipitated 3-phenylorotic acid monohydrate (1·25 g.) which separated from water as needles, m. p. and mixed m. p. 280° (decomp.).

5-*Carboxymethylene*-3-*phenylhydantoin.*—Dry hydrogen chloride was passed through a mixture of oxaloacetic acid (6·6 g.) and phenylurea (6·8 g.) in glacial acetic acid (50 ml.) for 30 min. at 80°. The solution was kept at 0° for 3 days, a solid separating. 5-*Carboxymethylene*-3-*phenylhydantoin* (2·1 g.), crystallised from water, had m. p. 200° (decomp.) (Found: C, 56·8; H, 3·45; N, 12·15. $C_{11}H_8O_4N_2$ requires C, 56·9; H, 3·45; N, 12·05%). Hydrolysis of the acid with aqueous potassium hydroxide gave 3-phenylorotic acid.

Decarboxylation of 3-*Phenylorotic Acid.*—A solution of 3-phenylorotic acid monohydrate (0·84 g.) in quinoline (4 ml.) was boiled with copper powder (0·1 g.) for 1 hr. The cooled solution was treated with N-sodium hydroxide (20 ml.) and extracted with ether. The alkaline solution was acidified and cooled overnight, crystals separating. 3-Phenyluracil (0·34 g.) separated from water as needles, m. p. and mixed m. p. 246—247° (Found: C, 63·8; H, 4·45; N, 14·7. Calc. for $C_{10}H_8O_2N_2$: C, 63·8; H, 4·45; N, 14·9%). A mixed m. p. with 1-phenyluracil (m. p. 246°) was 210°.

Decarboxylation of Orotic Acid.—Orotic acid (0·18 g.) in quinoline (3 ml.) containing copper powder (0·03 g.) was boiled under reflux for 1 hr. The cooled solution was treated with 2N-sodium hydroxide (4 ml.) and water (2 ml.) and extracted with ether. The aqueous phase was acidified and continuously extracted with ethyl acetate. Evaporation of the solvent gave uracil (0·05 g.) which separated from water as needles, m. p. and mixed m. p. 330° (decomp.) (Found: N, 24·9. Calc. for $C_4H_4O_2N_2$: N, 25·0%). The compound was indistinguishable from uracil in a paper chromatogram run in butanol, formic acid, water (77 : 13 : 10) (R_F = 0·32).

[9] Behrend and Struve, *Annalen*, 1911, **378**, 153.

Decarboxylation of 3-Methylorotic Acid.—5-Ethoxycarbonylmethylene-3-methylhydantoin was prepared from methylurea and diethyl oxaloacetate. The product had m. p. 135°. The hydantoin (1 g.) was heated with 2N-sodium hydroxide (10 ml.) for 30 min. at 100°. The cooled solution was acidified with 10N-hydrochloric acid, 3-methylorotic acid (0·7 g.), m. p. 300° (decomp.), being precipitated; this recrystallised from water (Found: C, 42·55; H, 3·55; N, 16·55. Calc. for $C_6H_6O_4N_2$: C, 42·35; H, 3·55; N, 16·45%). The acid was suspended in quinoline (4 ml.) containing copper powder (0·1 g.) and the mixture boiled for 45 min. The cooled solution was treated with 2N-sodium hydroxide (10 ml.) and extracted with ether. The aqueous phase was acidified and continuously extracted with ethyl acetate for 3 hr. Evaporation of the ester solution gave a solid residue. 3-Methyluracil (0·06 g.) separated from ethanol as needles, m. p. and mixed m. p. 179° (Found: N, 22·35. Calc. for $C_5H_6O_2N_2$: N, 22·2%).

3-p-*Sulphamoylphenylorotic Acid.*—*N*-Ethoxycarbonylaminomaleic anhydride (0·55 g.) and sulphanilamide (0·42 g.) were warmed together in ethanol (5 ml.) until a clear solution was obtained. The cooled solution gave a crystalline precipitate; α-N-*ethoxycarbonylamino*-N′-p-*sulphamoylphenylmaleimide* (0·3 g.) separated from ethanol as needles, m. p. 240° (decomp.) (Found: C, 45·9; H, 4·1; N, 12·2. $C_{13}H_{13}O_6N_3S$ requires C, 46·0; H, 3·9; N, 12·4%). The maleimide (0·6 g.) was dissolved in warm 2N-sodium hydroxide (3 ml.), and the solution acidified to give a solid precipitate. 3-p-*Sulphamoylphenylorotic acid* (0·28 g.) separated from water as hydrated needles, m. p. 320° (decomp.) (Found, in material dried at 100°/0·1 mm.: C, 41·9; H, 3·4; N, 13·15. $C_{11}H_9O_6N_3S,\frac{1}{4}H_2O$ requires C, 41·9; H, 3·3; N, 13·3%). Found, in material dried at 136°/0·1 mm.: C, 42·6; H, 3·2; N, 13·3. $C_{11}H_9O_6N_3S$ requires C, 42·4; H, 2·9; N, 13·5%).

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