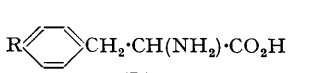
292. *Bacteriostasis in the Amino-acid Series. Part II. Further Studies with Alanine Derivatives.*

By D. F. Elliott and Sir Charles Harington.

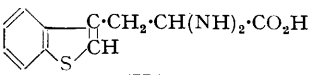
The search for antibacterial activity in the amino-acid group has been continued. The synthesis of several more alanine derivatives with widely different structures is described. One only of these amino-acids, β-3-thianaphthenylalanine, exerted a significant bacteriostatic action, and this was confined to *Strep. hæmolyticus*.

In continuation and extension of the work described in Part I (*J.*, 1948, 85) the present paper describes the synthesis of derivatives of β-phenylalanine substituted in the *p*-position with the following groups: guanidino, guanidinomethyl, *p*-aminophenylsulphonyl, and mercapto. The first two were selected in further development of the previous investigation of the effect of basic substituents in conferring bacteriostatic action on alanine derivatives; attempted extension of this line of work to the synthesis of *p*-amidino- and *p*-2′-diethylaminoethylamino-derivatives of β-phenylalanine was abandoned owing to preparative difficulties. The amino-phenylsulphonyl derivative seemed worth studying in view of the known tuberculostatic action of certain diaryl sulphones carrying basic substituents.

The *p*-mercaptophenylalanine (I; R = SH) was considered to be a possible tyrosine antagonist which might therefore inhibit the growth of micro-organisms with obligatory requirements for aromatic amino-acids. With a similar thought in mind, a possible inhibitor of tryptophan metabolism, namely β-3-*thianaphthenylalanine* (II) has been prepared and its synthesis is here described. Since our work was completed, a different synthesis of this amino-acid has been published by Avakian, Moss, and Martin (*J. Amer. Chem. Soc.*, 1948, **70**, 3075).



(I.)



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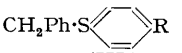

(III.)

β-p-*Guanidinophenylalanine* [I; R = NH·C(NH$_2$):NH] was prepared by the action of cyanamide on *ethyl acetamido-4-aminobenzylmalonate*, followed by acid hydrolysis of the product. The homologous guanidinomethyl derivative [I; R = ·CH$_2$·NH·C(NH$_2$):NH] was obtained by acid hydrolysis of the product of condensation of ethyl acetamido-4-aminomethylbenzylmalonate (Part I, *loc. cit.*) with *S*-methyl*iso*thiourea; difficulties were encountered in effecting this condensation until it was found that the reaction proceeded smoothly in phenol solution at 115°.

In the synthesis of β-p-(p′-*aminophenylsulphonyl*)*phenylalanine* (I; R=*p*-NH$_2$·C$_6$H$_4$·SO$_2$·), *p*-nitrophenyl *p*-tolyl sulphide (Law and Johnson, *J. Amer. Chem. Soc.*, 1930, **52**, 3625) was oxidised with hydrogen peroxide to give the corresponding *sulphone*. This sulphone was brominated in the ω-position, and the product was condensed with ethyl acetamidomalonate; the resulting *ester*, on hydrolysis with hydrobromic and acetic acid, was smoothly converted into the *amino-acid* (I; R = *p*-NO$_2$·C$_6$H$_4$·SO$_2$). Catalytic reduction then yielded the desired aminophenylsulphonylphenylalanine.

β-*p*-Mercaptophenylalanine has already been prepared by a somewhat cumbersome method

(Johnson and Brautlecht, *J. Biol. Chem.*, 1912, **12**, 175). The procedure now described offers advantages of ease of manipulation. The starting point of the synthesis was 4 : 4′-dicarboxydiphenyl disulphide; by reduction with sodium in liquid ammonia, followed by treatment with benzyl chloride, this compound was converted in one operation into p-*benzylthiobenzoic acid* (III; R = CO_2H). The derived *ethyl* ester was converted successively into the *hydrazide* and *toluene*-p-*sulphonhydrazide*; the sulphonhydrazide, under the conditions prescribed by McFadyen and Stevens (*J.*, 1936, 584), yielded p-*benzylthiobenzaldehyde* (III; R = CHO). The aldehyde was condensed with hippuric acid, and the resulting *oxazolone* hydrolysed to α-*benzamido*-p-*benzylthiocinnamic acid* [III; R = $CH{:}C(NHBz)\cdot CO_2H$]. On reductive hydrolysis, by boiling with hydriodic acid and phosphorus, the latter acid yielded the desired β-*p*-mercaptophenylalanine (I; R = SH). In the process of removal of hydriodic acid some oxidation to the disulphide invariably occurred; it was therefore convenient to complete this oxidation by means of iodine and to isolate the disulphide, from which the thiol could readily be obtained by reduction. The mercapto-amino-acid was obtained as the *hydrochloride dihydrate*, which was not described by Johnson and Brautlecht (*loc. cit.*).

For the synthesis of β-3-thianaphthenylalanine the starting point was thianaphthen-3-aldehyde. This aldehyde had been prepared in poor yield by Komppa and Weckmann (*J. pr. Chem.*, 1933, **138**, 109) by the action of ethyl orthoformate or ethoxymethyleneaniline on 3-thianaphthenylmagnesium bromide. We obtained moderate yields of the aldehyde both by the Rosenmund reduction of thianaphthen-3-carboxyl chloride and by the McFadyen–Stevens reduction of the corresponding *thianaphthen*-3-*carboxy*-p-*toluenesulphonhydrazide*. The aldehyde was condensed with hippuric acid in the usual way and the *oxazolone* reduced directly to *benzoyl*-β-3-*thianaphthenylalanine* by the method of Lamb and Robson (*Biochem. J.*, 1931, **25**, 1234). Hydrolysis with a mixture of hydrochloric and acetic acids gave the amino-acid (II).

Biological Results.—None of the above-mentioned amino-acids inhibited the growth of *S. aureus* or *E. coli* in broth up to the limit of their solubility. Against *Strept. hæmolyticus* in broth, (II) inhibited growth at a dilution of 1 in 20,000. The other amino-acids were very weakly active. We are indebted to Dr. A. T. Fuller for these results. Dr. P. D'Arcy Hart has also tested (I; R = p-$NH_2\cdot C_6H_4\cdot SO_2$) against *M. tuberculosis*; the compound has slight tuberculostatic action *in vitro*, but is less effective than *pp′*-diaminodiphenyl sulphone.

Experimental.

(M. p.s are uncorrected.)

Ethyl Acetamido-4-*nitrobenzylmalonate.*—To a solution of sodium (1·15 g.) in dry alcohol (25 c.c.) was added ethyl acetamidomalonate (10·9 g.), and, when the ester had dissolved, a hot solution of 4-nitrobenzyl bromide (11·3 g.) in alcohol (25 c.c.) was added in one portion. When the vigorous reaction had subsided, the mixture was boiled under reflux for 15 minutes and then diluted with water. The *ester* was filtered off, washed with water, and crystallised from alcohol. Yield, quantitative; m. p. 190° (Found: N, 7·95. $C_{16}H_{20}O_7N_2$ requires N, 8·0%).

Ethyl Acetamido-4-*aminobenzylmalonate.*—The above nitro-compound (3·5 g.) was suspended in alcohol (100 c.c.), Raney nickel (1 g.) was added, and the mixture hydrogenated at room temperature and pressure. Complete hydrogenation of the nitro-group required 6 hours owing to the sparing solubility of the nitro-compound. The solution was filtered and evaporated to dryness under reduced pressure, and the residue crystallised from benzene. It formed colourless prisms, m. p. 132°. Yield, 2·8 g. (87%) (Found: N, 8·8. $C_{16}H_{22}O_5N_2$ requires N, 8·7%).

Ethyl Acetamido-4-*guanidinobenzylmalonate Picrate.*—The amine (4·8 g.) and cyanamide (1·5 g.) were heated under reflux in alcohol (60 c.c.) containing one equivalent of hydrogen chloride for 8 hours. The solution was then evaporated to dryness under reduced pressure, and the residual oil lixiviated with cold water (*ca.* 30 c.c.) and filtered. A large excess of solid ammonium nitrate was added to the filtrate. Next day the crude nitrate was collected and dissolved in hot water (*ca.* 250 c.c.), and a hot aqueous solution of ammonium picrate added in excess. When cold, the *picrate* was filtered off and crystallised several times from alcohol. It formed rectangular plates, m. p. 190—191° (4·8 g.) (Found: N, 16·2. $C_{23}H_{27}O_{12}N_7$ requires N, 16·5%).

β-p-*Guanidinophenylalanine Monohydrochloride.*—The pure picrate described above (3 g.) was boiled under reflux for 4 hours with 3N-hydrochloric acid (20 c.c.). The picric acid was then removed by several extractions with warm benzene, and the aqueous layer evaporated to dryness under reduced pressure. The residual syrup was dissolved in 2 c.c. of hot water, and the solution neutralised to pH 4 with concentrated aqueous ammonia. The crude *monohydrochloride* (1·1 g.) which separated on cooling could not be obtained pure by recrystallisation. 1·3 G. were dissolved in hot water (15 c.c.), and a solution of ammonium picrate (1·5 g.) in hot water (15 c.c.) was added. When the solution cooled, a mixture of orange prisms and pale-yellow needles separated. The whole was then carefully reheated, with stirring, until the yellow needles had redissolved. The liquid was decanted, and the orange prisms remaining were recrystallised several times from hot water. The pure *monopicrate* had m. p. 226—228° (decomp.) (Found: C, 42·85; H, 3·7; N, 21·8. $C_{16}H_{17}O_9N_7$ requires C, 42·6; H, 3·8; N, 21·7%). The pure monohydrochloride was obtained from the monopicrate (1·45 g.) by adding it to 3N-hydrochloric acid (4 c.c.), extracting several times with warm benzene, evaporating it to dryness under reduced

pressure, redissolving the syrup in hot water (1 c.c.), and adding concentrated aqueous ammonia to pH 4, followed by alcohol (3 c.c.). The monohydrochloride separated in hexahedral crystals, m. p. 240—242° (decomp.) (Found : N, 21·0. $C_{10}H_{15}O_2N_4Cl$ requires N, 21·7%). The Sakaguchi and the ninhydrin reaction were positive.

Ethyl Acetamido-4-guanidinomethylbenzylmalonate Picrate.—Ethyl acetamido-4-aminomethylbenzylmalonate hydrochloride (Part I, *loc. cit.*; 14·8 g.) was added to one equivalent of ice-cold N-sodium hydroxide, the free base extracted with chloroform, and the chloroform extract dried and evaporated under reduced pressure without heating. The residue was dissolved in warm phenol (40 g.), *S*-methyl-*iso*thiourea sulphate (5·6 g.) added, and the mixture heated to 115°. The sulphate slowly dissolved with evolution of methylthiol. The mixture was occasionally shaken. After the mixture had been maintained for 2½ hours at 115—120°, the homogeneous solution was cooled, water (100 c.c.) added, and the phenol shaken out with ether. The lower layer was warmed under reduced pressure for a short while to remove ether, excess ammonium nitrate added, and the solution acidified with a few drops of dilute nitric acid. The crude nitrate which separated was filtered off and dissolved in hot water (200 c.c.), and a hot concentrated solution of ammonium picrate added until precipitation was complete. The crystalline solid was collected, washed by stirring with several small portions of hot water and filteng each time, and crystallised three times from alcohol. The pure picrate had m. p. 169—171°. Yield, 9·2 g. (38%) (Found : N, 15·9; OEt, 14·8. $C_{24}H_{29}O_{12}N_7$ requires N, 16·15; OEt, 14·8%).

β-p-*Guanidinomethylphenylalanine Monohydrochloride.*—The pure picrate described above (3 g.) was heated under reflux for 4 hours with 3N-hydrochloric acid (20 c.c.), and the solution extracted several times with warm benzene to remove picric acid. The aqueous layer was evaporated to dryness under reduced pressure, the residue dissolved in 5 c.c. of hot water, and the solution adjusted to pH 5 with concentrated ammonia solution. After 24 hours at 0° the *monohydrochloride* was filtered off and recrystallised from hot water. It formed rectangular plates, m. p. 290—292° (decomp.) [yield, 1·2 g. (88%)] (Found : C, 48·4; H, 6·3; N, 20·45. $C_{11}H_{17}O_2N_4Cl$ requires C, 48·4; H, 6·3; N, 20·5%). The Sakaguchi and the ninhydrin test were positive.

p-*Nitrophenyl* p-*Tolyl Sulphone.*—*p*-Nitrophenyl *p*-tolyl sulphide (Law and Johnson, *loc. cit.*; 19 g.) was dissolved with gentle warming in acetic acid (150 c.c.); the solution was treated with hydrogen peroxide (36 c.c. of 30%) which caused some precipitation; the mixture was cautiously warmed until reaction set in, and external heat was then withdrawn; after subsidence of the vigorous effervescence the solution was boiled for 2 hours and allowed to cool. The *sulphone* which separated (19·3 g.) was almost pure. After recrystallisation from acetic acid it formed stout pale yellow prisms, m. p. 176° (Found : S, 11·1. $C_{13}H_{11}O_4NS$ requires S, 11·5%).

p-*Nitrophenyl ω-Bromo*-p-*tolyl Sulphone.*—The preparation of this compound offered difficulty. With 1 mole of bromine a product containing only about 75% of the theoretical amount of bromine was obtained; on the other hand, if addition of bromine were carried further, some dibromination occurred. Best results were achieved by carrying out the reaction in two stages as follows. The sulphone (14 g.) was melted and mechanically stirred in an oil-bath at about 190°; bromine (8 g.) was added slowly, and the melt allowed to cool. Recrystallisation from acetic acid gave 14 g. of a product having m. p. 170° and containing 16% of bromine (theory, 22·5%). This product (12·85 g.) was again melted and treated with bromine in slight excess over the theoretical amount for completion of the reaction, the addition being interrupted when a fresh drop of bromine ceased to cause a brisk evolution of hydrogen bromide. Crystallisation of the reaction product first from acetic acid and then from methyl ethyl ketone yielded 8·7 g. of *bromo*-derivative having m. p. 174—176°. Even this sample was not quite pure, as shown by the analysis (Found : C, 44·7; H, 3·1; Br, 20·1. $C_{13}H_{10}O_4NSBr$ requires C, 43·8; H, 2·8; Br, 22·5%); it was, however, satisfactory for further reaction.

Ethyl Acetamido-p-(p′-*nitrophenylsulphonyl*)*benzylmalonate.*—Ethyl acetamidomalonate (4 g.) was dissolved in a solution of sodium (0·42 g.) in anhydrous alcohol (20 c.c.); the bromo-sulphone (6·5 g.) was dissolved in anhydrous dioxan (20 c.c.), and the solutions were mixed; after refluxing for 3½ hours reaction was complete; the mixture was cooled, diluted with water, and the *ester* collected and crystallised from alcohol. It formed colourless narrow prisms, m. p. 196° [6·63 g. (74%)] (Found : C, 53·3; H, 4·9; N, 6·0. $C_{22}H_{24}O_9N_2S$ requires C, 53·7; H, 4·9; N, 5·7%).

β-p-(p′-*Nitrophenylsulphonyl*)*phenylalanine.*—The preceding ester (6 g.) was dissolved in acetic acid (30 c.c.); hydrobromic acid (30 c.c., *d* 1·7) was added, and the mixture heated under reflux for 4 hours. On cooling, the hydrobromide of the amino-acid separated; it was collected and dried. Yield, 5 g. (95%). The free *amino-acid* was obtained by dissolving the hydrobromide in hot water with addition of hydrobromic acid, and neutralising at the boiling point with aqueous ammonia. It separated in bunches of colourless stumpy needles, m. p. 236—238° (decomp.) (Found : N, 7·6; S, 9·5. $C_{15}H_{14}O_6N_2S$ requires N, 8·0; S, 9·6%).

β-p-(p′-*Aminophenylsulphonyl*)*phenylalanine.*—The hydrobromide of the nitrophenylsulphonylphenylalanine (5 g.) was suspended in 50% alcohol (200 c.c.) to which was added hydrobromic acid (1·35 c.c., *d* 1·49; 1 equivalent). The mixture was hydrogenated at atmospheric pressure in presence of platinum oxide (150 mg.). The uptake of hydrogen became very slow after 3 hours, the reaction being still incomplete. Hydrobromic acid (5 c.c., *d* 1·49) and platinum oxide (100 mg.) were added, and hydrogenation resumed; the uptake finally (about 5 hours in all) ceased at 820 c.c. (theory at 23°/762 mm. is 850 c.c.). The solution was filtered and evaporated to dryness under reduced pressure. The residue was taken up in water, freed from a little black amorphous material by filtration, and neutralised to pH 7—8 at the boiling point by addition of aqueous ammonia. After keeping overnight at 0° the *amino-acid* was collected, washed with water, and dried. Yield, 3·1 g. (83%) of practically pure material. For analysis it was recrystallised by dissolution in water with the aid of ammonia and neutralisation to litmus at the boiling point with acetic acid. It formed colourless plates, m. p. 267—269° (decomp.), and was much more soluble in water than the corresponding nitro-compound (Found : C, 56·2; H, 5·3; N, 8·6; S, 9·8. $C_{15}H_{16}O_4N_2S$ requires C, 56·3; H, 5·0; N, 8·8; S, 10·0%).

p-*Benzylthiobenzoic Acid.*—*pp′*-Dicarboxydiphenyl disulphide was prepared from *p*-aminobenzoic acid according to method described in *Org. Synth.*, Coll. Vol. II, p. 580 for the *oo′*-isomer and was used

in the crude state, containing some salt. An amount of the crude material containing 15 g. of the disulphide was dissolved in liquid ammonia (300 c.c.), and sodium was added in small portions to the well-stirred solution until present in excess. Somewhat more than six atoms were needed. The blue colour caused by excess of sodium was discharged by addition of the minimum amount of ammonium chloride, and benzyl chloride (12·5 c.c.; 50% excess) was stirred in. The ammonia was allowed to evaporate, the residue dissolved in boiling water, and the solution filtered. After the mixture had been kept overnight at 0°, the sodium salt which had separated was collected, dissolved in boiling water, and acidified while hot with hydrochloric acid. The precipitated *acid* [16 g. (66%)] was collected after cooling and dried. It crystallised from acetic acid in colourless prisms, m. p. 189—190° (Found : S, 13·2. $C_{14}H_{12}O_2S$ requires S, 13·1%). The *ethyl* ester was prepared by boiling a solution of the acid (6·62 g.) in alcohol (70 c.c.) containing sulphuric acid (7 c.c.) for 4 hours, and pouring the hot solution into water (500 c.c.) containing sodium bicarbonate (22 g.); the ester was isolated by extraction with ether. Yield, 7 g. (95%). It formed colourless prisms from alcohol, m. p. 60° (Found : S, 12·0. $C_{16}H_{16}O_2S$ requires S, 11·8%).

p-*Benzylthiobenzhydrazide*.—The preceding ethyl ester (5·44 g.) was dissolved in alcohol (10 c.c.) containing hydrazine hydrate (1·5 c.c. of 100%) and the solution heated on the steam-bath in a pressure-bottle for 17 hours; the contents were poured whilst hot into a beaker, and the *hydrazide* which separated on cooling was collected (yield, 4·17 g.; 80%). Recrystallised from alcohol it formed colourless plates, m. p. 166° (Found : N, 11·2; S, 12·7. $C_{14}H_{14}ON_2S$ requires N, 10·9; S, 12·4%). The *toluene*-p-*sulphonyl* derivative was prepared by adding toluene-*p*-sulphonyl chloride (9·7 g.; 10% excess) in small portions to a solution of the hydrazide (11·8 g.) in dry pyridine (120 c.c.) with just enough warming to maintain a clear solution; after heating for 10 minutes on the steam-bath the mixture was poured into ice-water containing excess of hydrochloric acid, and the product collected and crystallised from acetic acid. Yield, 18·2 g. (96%). M. p. 180° (Found : N, 6·8; S, 15·5. $C_{21}H_{20}O_3N_2S_2$ requires N, 6·8; S, 15·4%).

p-*Benzylthiobenzaldehyde*.—The toluene-*p*-sulphonylhydrazide (18 g.) was heated in a bath at 160° with ethylene glycol (95 c.c.). The hot solution was treated with finely powdered anhydrous sodium carbonate (8·64 g.), heating being continued for 1 minute. The solution was slightly cooled and diluted with hot water (1 l.). After rapid cooling, the oil was isolated with the aid of ether; the residue after evaporation of the solvent was entirely crystalline and practically pure *aldehyde* (8 g.; 80%). For analysis a small sample was distilled under reduced pressure and crystallised from alcohol; it formed very pale-yellow leaflets, m. p. 70° (Found : C, 74·0; H, 5·6; S, 14·0. $C_{14}H_{12}OS$ requires C, 73·7; H, 5·3; S, 14·0%).

2-*Phenyl*-4-(4′-*benzylthiobenzylidene*)*oxazol*-5-*one*.—The aldehyde (8 g.), fused sodium acetate (8 g.), and hippuric acid (6·3 g.) were intimately mixed, treated with acetic anhydride (50 c.c.), and heated on the steam-bath for 15 minutes. Complete dissolution occurred before the product began to crystallise. After cooling, the product was stirred with water containing some sulphuric acid to assist the decomposition of excess acetic anhydride; the *oxazolone* was then collected, washed with water, and crystallised from acetic acid. Yield, 8·5 g. (74%) of orange prisms, m. p. 186° (Found : N, 3·9; S, 8·7. $C_{23}H_{17}O_2NS$ requires N, 3·8; S, 8·6%).

α-*Benzamido*-4-*benzylthiocinnamic Acid*.—The preceding oxazolone (14·9 g.) was added to boiling 66% alcohol (1490 c.c.) containing 1% of sodium hydroxide. The solution cleared in 1—2 minutes, and boiling was continued for about 10 minutes. Dilute hydrochloric acid was then added in excess, followed by enough water to produce turbidity. On cooling the pure *acid* separated in long prismatic needles (14·75 g.; 96%), m. p. 21—214°, unchanged by recrystallisation from 65% alcohol (Found : N, 3·9. $C_{23}H_{19}O_3NS$ requires N, 3·6%).

β-p-*Mercaptophenylalanine*.—The benzamidocinnamic acid (9·8 g.), red phosphorus (10 g.), and hydriodic acid (200 c.c., *d* 1·7) were boiled under reflux for 3 hours. After cooling the solution was filtered through asbestos, and the filtrate evaporated to dryness under reduced pressure. The dark residue immediately became colourless on treatment with water, indicating oxidation of the thiol compound by residual free iodine; the aqueous solution was therefore extracted with ether to remove benzoic acid, freed from ether by distillation, chilled in ice, and treated with 4N-iodine in potassium iodide until free iodine was present in very slight excess; the mixture was then heated, and the hot solution brought to pH 5 by addition of aqueous ammonia. The disulphido-amino-acid separated in bunches of minute needles; when cold, it was collected, washed with alcohol and ether, and dried. Yield, 4·4 g. (88%). Purification of the disulphide was difficult; it was achieved with considerable loss as follows. 1·2 G. were dissolved by warming to 50° with N-hydrochloric acid (100 c.c.), and the yellow solution was allowed to stand overnight at 15°. The gelatinous precipitate was filtered off, and the filtrate neutralised at the boiling point with concentrated ammonia solution. After several hours the solid was filtered off, washed thoroughly with water, and dried at 100°/20 mm. for analysis. Yield, 0·42 g.; m. p. 258—260° (decomp.) (Found : C, 55·0; H, 5·7; N, 7·3; S, 15·5. Calc. for $C_{18}H_{20}O_4N_2S_2$: C, 55·1; H, 5·1; N, 7·1; S, 16·3%). For the preparation of the mercapto-amino-acid, the impure disulphide was partly dissolved in boiling 5N-hydrochloric acid (10 c.c.), and zinc dust (1 g.) was added in portions to the hot solution during 10—15 minutes. The solution was filtered and mixed at the boiling point with an equal volume of concentrated hydrochloric acid. On cooling, the *hydrochloride dihydrate* separated in bunches of tiny needles which sintered and darkened at 200° and decomposed at 222° [Found : N, 5·2; S, 11·7; SH (by iodine titration) 12·2. $C_9H_{16}O_4NClS$ requires N, 5·2; S, 11·9; SH, 12·25%].

Thianaphthen-3-*aldehyde*.—(*a*) Thianaphthen-3-carboxyl chloride (Crook and Davies, *J.*, 1937, 1697; 6·3 g.) was dissolved in dry xylene (30 c.c.), 5% palladised barium sulphate (3 g.) added, and the mixture heated under reflux with vigorous stirring in a stream of hydrogen. After 4 hours, evolution of hydrogen chloride had practically ceased and the reduction was discontinued. The solution was filtered, and the filtrate shaken for 15 minutes with excess of saturated sodium bisulphite solution. The bisulphite compound was filtered off, washed with xylene, and dried on a tile. The pure aldehyde was obtained on decomposition with warm sodium carbonate in the usual way. Yield, 2·2 g. (42·5%); m. p. 54°.

(*b*) Methyl thianaphthen-3-carboxylate (Komppa and Weckmann, *loc. cit.*; 15 g.) was boiled under reflux for 8 hours with alcohol (37·5 c.c.) and hydrazine hydrate (9 c.c. of 90%). Water was then added to the solid mass, and the *thianaphthen-3-carboxyhydrazide* filtered off and crystallised from 30% methanol. It formed sheaves of needles, m. p. 176—177° (12·2 g.; 68%) (Found : C, 55·9; H, 4·3; N, 15·2. $C_9H_8ON_2S$ requires C, 56·25; H, 4·2; N, 14·6%). The hydrazide (11·3 g.) was dissolved in dry pyridine (192 c.c.), and the solution cooled to 0°. Toluene-*p*-sulphonyl chloride (11·3 g.) was then added in small portions during 15—20 minutes. After being kept at room temperature for 2 hours, the mixture was poured into 5N-HCl (465 c.c.) with stirring. The solid was collected and crystallised from 80% acetic acid. The yield of *thianaphthen-3-carboxytoluene-p-sulphonylhydrazide* was 19·2 g. (94%); m. p. 201—203°. It was dried at 110° for analysis (Found : C, 55·6; H, 4·0; N, 8·3. $C_{16}H_{14}O_3N_2S_2$ requires C, 55·5; H, 4·1; N, 8·1%). The toluene-*p*-sulphonyl derivative (3·1 g.) was dissolved in ethylene glycol (redistilled; 15 c.c.) and heated to 160°, and anhydrous sodium carbonate (0·96 g.) added in one portion. When the vigorous effervescence had ceased, heating was continued for 30 seconds, and hot water (150 c.c.) added. The aldehyde was extracted with ether and purified through the bisulphite compound as before. Yield, 0·9 g. (63%).

2-*Phenyl-4-3′-thianaphthenylmethyleneoxazol-5-one*.—The aldehyde (0·7 g.), sodium hippurate (1 g.), and acetic anhydride (2·5 c.c.) were mixed and heated at 100° for 30 minutes. The resultant yellow mass was cooled, rubbed with water, and filtered. The *oxazolone* crystallised from benzene in long yellow needles, m. p. 219—220° (Found : N, 4·8. $C_{18}H_{11}O_2NS$ requires N, 4·8%).

N-*Benzoyl-β-3-thianaphthenylalanine*.—The oxazolone (0·8 g.) was added to a mixture of pure acetic acid (4·5 c.c.), hydriodic acid (0·1 c.c.; *d* 1·7), and red phosphorus (0·2 g.) and heated under reflux for 1 hour. The solution was filtered hot, and the phosphorus washed with hot acetic acid. The *benzoyl* compound crystallised from the filtrate on cooling and addition of water. It was recrystallised from glacial acetic acid, forming hexagonal plates, m. p. 226—228° (0·7 g.; 87%) (Found : C, 66·1; H, 4·4. $C_{18}H_{15}O_3NS$ requires C, 66·4; H, 4·6%). Thianaphthen was unchanged when subjected to the above treatment, showing that under these conditions the double bond in the heterocyclic ring is not reduced.

β-3-Thianaphthenylalanine.—The benzoyl derivative (1·5 g.) was heated under reflux with a mixture of concentrated hydrochloric (50 c.c.) and glacial acetic acid (20 c.c.) for 24 hours. The light-brown solution was treated with charcoal and evaporated to dryness under reduced pressure. The residue was taken up in hot water (15 c.c.), and the solution adjusted to pH 5 with concentrated ammonia solution. The *amino-acid* separated in small plates, m. p. 248—250° (decomp.) (Found : C, 59·5; H, 5·4. $C_{11}H_{11}O_2NS$ requires C, 59·7; H, 5·0%).

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