It can be concluded that substitution in position 4 ($R_1 = CH_3$) increases the anti-convulsant activity of the parent compound whereas substitution in position 1 yields less active products. The activity is further enhanced by halogenation of the benzene nucleus.

All of the compounds (X–XI) showed a low acute toxicity on oral administration. An exception was presented by the last two compounds (XII and XIII).

The pharmacological and toxicological properties of these compounds seem to justify clinical trials. The pharmacological features of these compounds will be described extensively elsewhere.

Riassunto. Sono stati studiati 13 derivati della 3,1-benzossazina, la maggior parte dei quali sono risultati attivi come anticonvulsivanti. Tra di essi il più interessante è risultato il 4H-4-dimetil-6-bromo-3,1-benzossazine-2-one (IV).

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An Azomethine-Hypophosphorous Acid Addition Compound as a new Colorimetric Reagent for Iron

For studies of tumour-inhibitory activity we prepared N,N'-*bis*-salicylidene derivatives of 1,3-diaminopropane and 1,2-diaminoethane[1] and the N-salicylidene compound of D-(+)-glucosamine[2]. Phosphonous acid derivatives were obtained from the azomethines by a general reaction: $o\text{-}HOC_6H_4CH = NR + H_3PO_2 \rightarrow o\text{-}HOC_6H_4CH(PO_2H_2)\text{-}NHR$ first described by Schmidt[3].

By chance it was found that aqueous solutions of the phosphonous acids gave stable red colours with ferric iron and the 1,2-diaminoethane compound proved to be most sensitive. Here we describe the preparation of this derivative and its use for the colorimetric determination of iron.

Materials, methods and results. N,N'-*bis*-salicylidene-1,2-diaminoethane phosphonous acid derivative [$o\text{-}HOC_6H_4CH(PO_2H_2)NH(CH_2)_2NHCH(PO_2H_2)C_6H_4OH\text{-}o$, SDEP].

N,N'-*bis*-salicylidene-1,2-diaminoethane (2.68 g; mp 127°) was dissolved in hot absolute ethanol (50 ml) and a slight excess of anhydrous hypophosphorous acid (~ 1.499; > 2 mole proportions) was added. Soon the yellow colour of the azomethine disappeared and a white precipitate formed. The mixture was refluxed for 10 min and kept overnight at room temperature. Precipitation of the phosphonous acid derivative was completed by addition of several volumes of anhydrous ether. SDEP was collected, washed with anhydrous ether and dried in air. Yield = 4 g; mp 206–207° with gas evolution. Microanalysis showed that the compound contained water. A sample was dried at 60° in vacuo for 12 h and then analyzed. During weighing, the analytical sample gained several µg.

For $C_{16}H_{22}N_2P_2O_6 \cdot {}^1/_2H_2O$,

calcd. C% = 46.95; H% = 5.69; N% = 6.84
found C% = 46.77; H% = 5.63; N% = 7.17

Colour reaction of ferric iron with SDEP. Aliquots of a freshly prepared solution of ferric alum (analytical reagent grade: 0.4012 g in 100 ml of deionized water) were added to 5 ml portions of 0.4% (w/v) aqueous solution (pH ~3) of SDEP to give concentrations of ferric iron ranging from 0 to 15 µg/ml. Absorption spectra were determined against water by means of a Unicam SP 800 recording spectrophotometer. The coloured complex gave a prominent absorption maximum at 495 nm. At a concentration of 15 µg of Fe^{+++}/ml E was ~0.94. Beer's law was obeyed.

Over the pH range 3–8, E_{495} was practically constant. The colour was partially discharged when the pH was brought to 2 by hydrochloric acid but was restored when the pH was increased to 3 or over by addition of sodium hydroxide solution.

We examined the effect of various concentrations of SDEP (0.05–0.4 g/100 ml) on E_{495} of solutions containing 11 µg of Fe^{+++}/ml and found that E_{495} was not markedly influenced by SDEP concentration.

Colorimetric determination of ferric iron directly and of ferrous iron (after hydrogen peroxide treatment) with SDEP reagent at 495 nm

Iron content (µg/ml)	I 0.4% SDEP + ferric E ± S.D.	II 0.1% SDEP + ferric E ± S.D.	III 0.1% SDEP + ferrous E	IV 0.1% SDEP + ferrous + H_2O_2 E ± S.D.	V 0.1% SDEP + ferric + ferrous E	VI 0.1% SDEP + ferric + ferrous + H_2O_2 E
1.9	0.142 ± 0.100	0.127 ± 0.018	0.004	0.131 ± 0.005	0.075	0.158
3.9	0.259 ± 0.006	0.276 ± 0.010	0.004	0.281 ± 0.006	0.144	0.274
7.7	0.514 ± 0.012	0.541 ± 0.014	0.000	0.537 ± 0.011	0.283	0.545
11.4	0.736 ± 0.011	0.781 ± 0.013	0.002	0.786 ± 0.010	0.399	0.784
15.1	0.942 ± 0.015	1.034 ± 0.012	0.001	1.037 ± 0.005	0.520	1.042

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Calibration curves were prepared for ferric iron by means of a Unicam SP 600 spectrophotometer and in the Table are given E_{495} ± standard deviation (S.D.) for 3 sets of curves in 0.4% (Column I) and 0.1% (Column II) SDEP solutions.

These readings were made 30 min after preparation of the solutions. The colours were stable for at least 24 h. At the higher concentrations of Fe^{+++}, E values were somewhat greater in 0.1% than in 0.4% SDEP reagent.

Behaviour of ferrous iron in 0.1% SDEP. Aliquots of a freshly prepared solution of ferrous alum (analytical reagent grade: 0.3263 g in 100 ml of deionized water) were added to 5 ml portions of 0.1% SDEP. No colour was detected in any tube 30 min after preparation (Table, column III). To each tube was added 0.01 ml of 30% hydrogen peroxide and the colours read at 495 nm after 30 min. Results for 3 sets of experiments are given in the Table, column IV. They are practically identical with those obtained in the direct reaction of SDEP with ferric iron (Column II).

Determination of ferric and ferrous iron in the same solution. A stock solution (100 ml) of 0.2006 g of ferric alum and 0.1632 g of ferrous alum was prepared and aliquots of the fresh solution which contained equal parts of ferric and ferrous iron, were added to 5 ml portions of 0.1% SDEP to give final concentrations of 1.9–15.1 µg of iron/ml. Colours were read after 30 min (Table, column V) and again (Table, column VI) 30 min after addition of 0.01 ml of 30% hydrogen peroxide to each tube. E_{495} for each peroxide-treated solution was approximately twice that of the untreated solution and the final readings were the same as those given by solutions containing Fe^{+++} only (Table, column II). Thus the direct reaction with SDEP is capable of determining ferric iron in a mixture of ferric and ferrous iron, and after peroxide addition, the increase in E_{495} permits determination of the ferrous iron content of the mixture.

Discussion. SDEP solutions are clear and colourless and seem to be stable indefinitely at room temperature. In this respect, the reagent is superior to a similar compound ethylenediaminedi(*o*-hydroxyphenylacetic acid) o–$HOC_6H_4CH(COOH)NH(CH_2)_2NHCH(COOH)C_6H_4OH$–o which was investigated by UNDERWOOD[4] as a possible reagent for ferric iron.

None of the common metal ions such as Cu^{++}, Mn^{++} and Ni^{++} gave a colour with SDEP reagent, but a pale green colour was given by K^{+}[4].

Résumé. Nous avons préparé un nouveau produit par réaction entre le N,N'-salicylidène-1,2-diaminoéthane et l'acide hypophosphoreux. Ce composé donne un complexe coloré avec l'ion ferrique et il semble convenir au dosage colorimétrique du fer.

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[5] Our thanks are due to Miss M. A. MCKINNON, Department of Chemistry, University of Sheffield, who carried out the microanalyses.

Synthesis of (±)-Isoelliptone

Isoelliptone[1] (erosone)[2], $C_{20}H_{16}O_6$, has been isolated along with some rotenoids (rotenone, pachyrrhizone etc.) from yam beans (*Pachyrrhizus erosus*). Its structure was shown to be an isomer of elliptone (I)[3] by NORTON and HANSBERRY[2], and lately was recognized as II on the basis of spectral studies by OLLIS et al.[1,4]. In previous papers[5,6], the authors have reported a convenient synthetic method of rotenoids (munduserone and elliptone) from corresponding 2'-hydroxyisoflavone derivatives[7,8]. The present paper will describe the synthesis of II from 2'-hydroxy-4',5'-dimethoxyfurano [3'',2'':6,7] isoflavone

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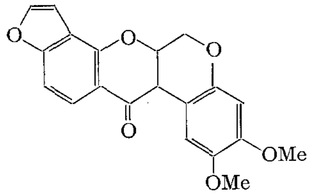
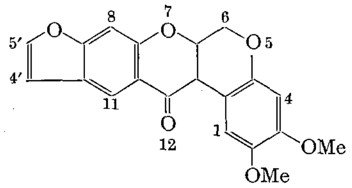
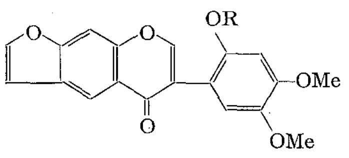


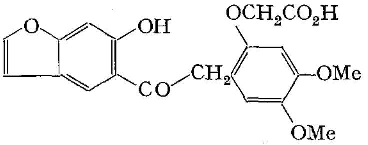
I



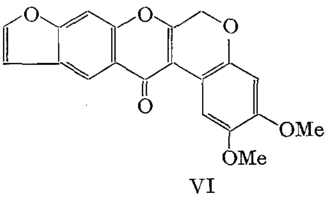
II



III R = H
IV R = $CH_2CO_2C_2H_5$



V



VI