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Reaction of Some Diketones with 5-Aminolevulinic Acid in Acid Solution

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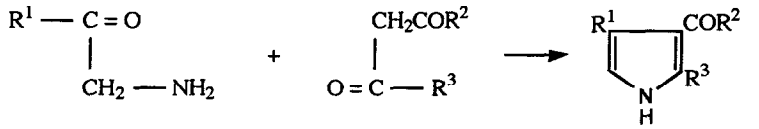
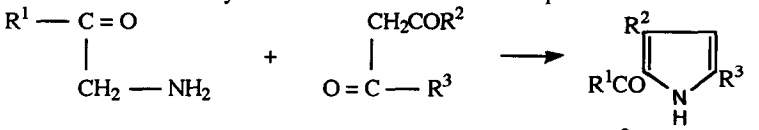
Abstract: Some 2,4-diketones react with 5-aminolevulinic acid in acid solution to form pyrroles There are two modes of cyclisation, one leading to the Knorr and the other to the Fischer-Fink product. By a consideration of the products obtained from pentane-, hexane-, 3-methylpentane-, 3-isopropylpentane-, 3,3-dimethylpentane-, 1,1,1-trifluoropentane- 1,1,1,5,5,5-hexafluoro- pentane-2,4-dione and ethyl acetoacetate some generalisations concerning the formation of Knorr and Fischer-Fink products have been made The reactions of 5-methyl-5-aminolevulinic acid with pentane- and 3-methylpentane-2,4-dione have also been examined. The relevance of these studies to the cyclic dimerisation of 5-aminolevulinic acid is discussed briefly

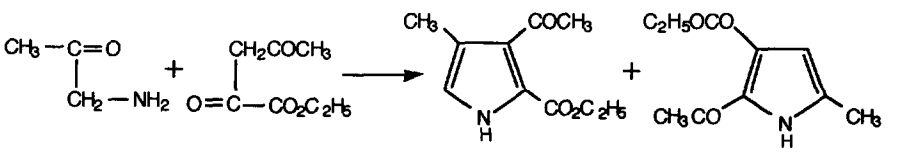
A regular route for the laboratory synthesis of pyrroles is the condensation of an α-aminoketone with a carbonyl compound containing an adjacent active methylene group. This route is generally known as the Knorr



synthesis.[1] If the methylene group is activated by a second carbonyl group there is an alternative mode of cyclisation known as the Fischer-Fink synthesis.[2] In some instances both products are obtained but not in equal



amounts, e.g. in the reaction of α-aminoacetone (**1**) with ethyl oxalylacetone (**2**),[3] **3** is the Knorr product while **4** is the Fischer-Fink product. The relative amounts of **3** and **4** depend upon the pH (Scheme 1).

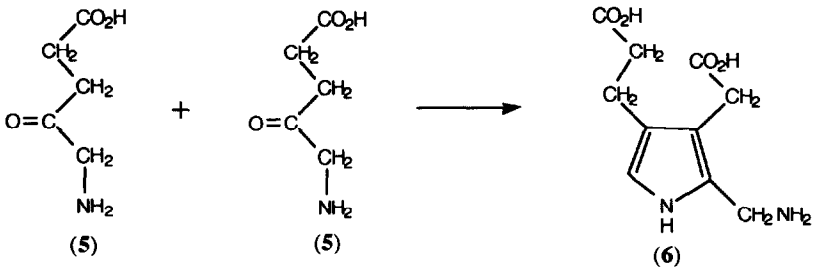


(**1**) (**2**) (**3**) (**4**)

SCHEME 1

In most cyclisation reactions of this type the Knorr product is the dominant one. We have explored the situation with respect to one α-aminoketone, 5-aminolevulinic acid (5-ALA; **5**). Our interest arose because of

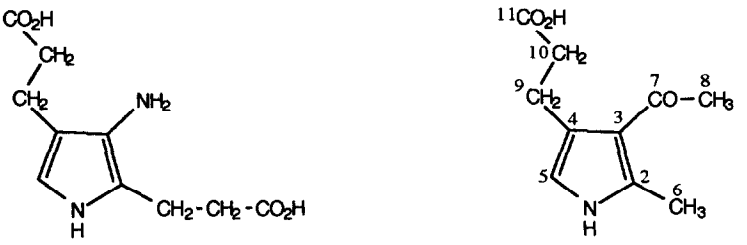
the important enzyme-catalysed reaction in which two molecules of 5-ALA condense to give porphobilinogen (**6**), an important building block in the biosynthesis of porphyrins, chlorophyll and corrins (Scheme 2). This is



(**5**) (**5**) (**6**)

SCHEME 2

the Knorr product but cyclisation *via* the 5-methylene group of the second molecule of **5** is possible to give *pseudo*porphobilinogen (**7**).



(**7**) (**8**)

The mechanism of the enzyme-catalysed process has been examined by a number of workers but most recently by Jaffe and Markham.[4,5] They have shown that 5-ALA forms a Schiff base with the enzyme (5-aminolevulinate dehydratase, more appropriately known as porphobilinogen synthase) and that an imine structure is the predominant tautomeric form of the Schiff base. They have also explored the protonation states of the two nitrogens in the first Schiff base complex.[6]

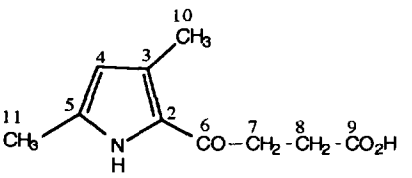
In the absence of enzyme no pyrrole is formed and so, for model studies, we have taken a related reaction, that between 5-ALA and pentane-2,4-dione (acetylacetone). This has allowed us to explore the process of acid catalysis occurring in the cyclisation to give a pyrrole and details of this will be published subsequently. First we need to identify the products of reaction. The reaction of pentane-2,4-dione with 5-ALA has long been used as an assay for 5-ALA in biological fluids,[7] particularly for the detection of lead poisoning which inhibits the conversion of 5-ALA into PBG. The product of reaction is generally taken to be 3-acetyl-4-(2-carboxyethyl)-2-methylpyrrole (**8**) and the quantitative determination of **8** is based on its reaction with Ehrlich's reagent (N,N-dimethylaminobenzaldehyde in acid solution) to give a purple adduct.[8] However we have found that **8** is not the only product and further studies of related reactions have provided insight into factors controlling the mode of cyclisation (Knorr and Fischer-Fink). The use of high field NMR spectroscopy has allowed us to detect even minor components in the product without tedious and uncertain separation procedures.

Any deduction from our results in terms of a mechanism of cyclisation depends upon a knowledge of the form adopted by 5-ALA in aqueous solution. This has been studied, using a number of isotopic probes (^{13}C NMR spectroscopy of [4-^{13}C]ALA, ^{1}H NMR spectroscopy, $^{1}H/^{2}H$ and $^{16}O/^{18}O$ exchange) by Jaffe and Rajagopalan.[9] Repetition of their work has been described in a previous publication[10] but reference is best made to the original report Studies of $^{1}H/^{2}H$ exchange demonstrated that enols are formed at both C_5 and C_3 and that the former occurs more readily. More extensive work by Jaffe and Rajagopalan[9] showed that at pH 6 8 enolisation at C_5 is 5 times faster than at C_3 and that in more acid solution, relevant to our work, the rate is

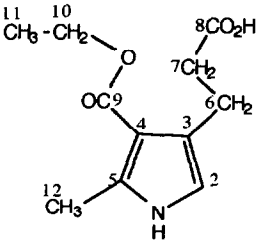
slower but the ratio is similar. In summary, Jaffe and co-workers have established that 5-ALA, in aqueous solution, can exist as a hydrate and in two enolic forms as well as the keto-compound

RESULTS

Reaction of 5-ALA with pentane-2,4-dione. In view of the low solubility of pyrrole compounds in water we can be sure that essentially all the pyrrolic products of reaction were contained in the precipitate formed as the reaction proceeded. Analysis of the product by NMR spectroscopy and by GC-MS revealed that it was a mixture of two products of identical relative molecular mass. The major product of reaction, constituting 95% of the total product, was the expected Knorr product **8**. The minor product, constituting 5% of the total product, was the Fischer-Fink product, 2-(3-carboxypropanoyl)-3,5-dimethylpyrrole (**9**). The ^{13}C NMR



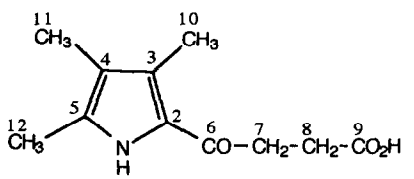
(**9**)



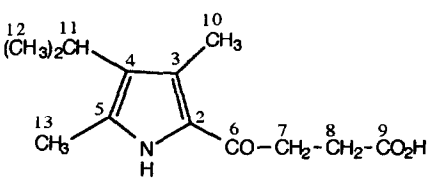
(**10**)

assignment of the pyrrole carbons in **8** (see Experimental) were confirmed by examination of material labelled with 50% enriched ^{13}C at the 4-position (made from enriched 5-ALA). Both the C_3 and C_5 resonances had satellite peaks on either side of the parent signal ($^1J_{C4C5}$ = 69.6 Hz; $^1J_{C3C4}$ = 53.7 Hz) and C_2 was assigned by default. Similarly the C_9 resonance had satellite peaks on either side of the parent signal ($^1J_{C4C9}$ = 48.8 Hz) and C_{10} was assigned by default. In the case of **9** with the ^{13}C label at C_6, satellite peaks were observed on either side of the C_7 resonance ($^1J_{C6H7}$ = 41.5 Hz) and C_8 was assigned by default. Because of the very small intensities of the pyrrole carbons, satellite peaks were not observed on either side of the C_2 resonance. The resonances of C_2, C_3, C_5, C_{10} and C_{11} of **9** were assigned by comparison with ^{13}C resonances of similar pyrroles[11]. The ^1H NMR assignments for both **8** and **9** were made from ^{13}C-^1H correlations.

Reaction of 5-ALA with ethyl acetoacetate. Only one solid product was obtained in this reaction and all the NMR spectra were consistent with its being the Knorr product 3-(2-carboxyethyl)-4-ethoxycarbonyl-5-methylpyrrole (**10**).



(**11**)



(**12**)

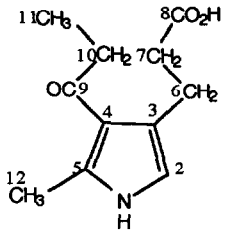
Reaction of 5-ALA with 3-methylpentant-2,4-dione. Only one solid product was obtained in this reaction and all the NMR spectra were consistent with its being the Fischer-Fink product, 2-(3-carboxypropanoyl)-3,4,5-trimethylpyrrole (**11**).

Reaction of 5-ALA with 3-isopropylpentane-2,4-dione. Analysis of the product by NMR spectroscopy revealed that only the Fischer-Fink product, 2-(3-carboxypropanoyl)-3,5-dimethyl-4-isopropylpyrrole (**12**) was formed in this reaction

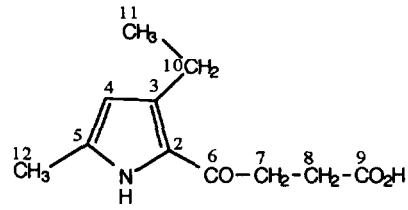
Reaction of 5-ALA with 3,3-dimethylpentant-2,4-dione. In spite of an extended time of reaction 3,3-dimethylpentane-2,4-dione was recovered unchanged. The only new material observed was 2,5-bis(2-carboxyethyl)-

dihydropyrazine, formed by the dimerisation of 2 molecules of 5-ALA (see later). A similar lack of reaction was observed by Combes.[12] The most likely explanation is that reaction of a dione with the amino group of 5-ALA requires formation of an enol in the former and this dione exists exclusively as the keto compound.

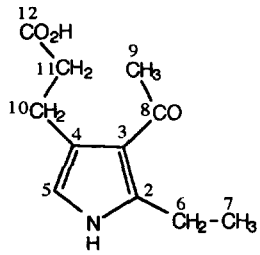
Reaction of 5-ALA with hexane-2,4-dione. Analysis of the product by both GC-MS and NMR spectroscopy revealed it to be a mixture of four products of identical relative molecular masses. Condensation of the amino group of 5-ALA can occur with either of the keto groups of this unsymmetrical dione but reaction at the 2-position is preferred in a ratio 2:1. Both intermediates can then cyclise by either the Knorr [(**13**) and (**15**)] or the Fischer-Fink [(**14**) and (**16**)] routes to give pyrrole products, with the former being dominant (19:1) in both cases.



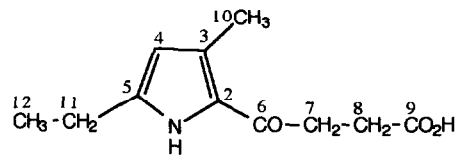
(**13**) (63%)



(**14**) (3%)

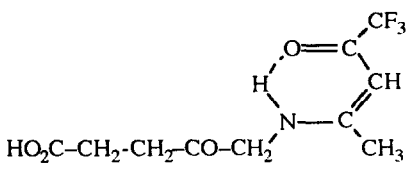


(**15**) (32%)

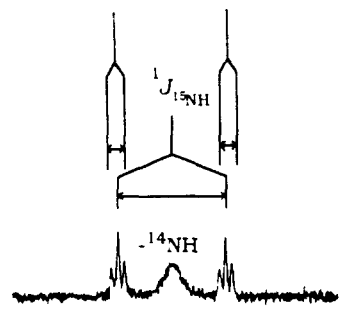


(**16**) (2%)

Reaction of 5-ALA with 1,1,1-trifluoropentant-2,4-dione. On reaction at room temperature, the product to come out of solution was not a pyrrole but the intermediate enaminoketone (**17**). The presence of the hydrogen bonded chelate ring was inferred from the large paramagnetic shift (δ 11.3) of the N-H proton. The structure of



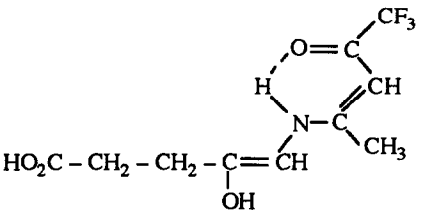
(**17**)



Figure

the enaminoketone was further confirmed from the following evidence. The low proton signal of the enaminoketone 50% enriched in ^{15}N appeared as a broad resonance at δ 11.3 (the ^{14}N compound) and a doublet on either side due to one bond ^{15}N-H coupling ($^{1}J_{^{15}NH}$ = 92.5 Hz) (Figure). Each signal of the doublet was further split into a triplet due to coupling with the protons of the α-CH_2 group attached to the nitrogen atom ($^{3}J_{HH}$ = 5.47 Hz). The splitting of the α-CH_2 protons into a doublet due to coupling with the NH protons, however, was not observed.

Further evidence that the product was an enaminoketone, and not an imine, was obtained from the ^{15}N chemical shift (δ 120.5) of the product 50% enriched in ^{15}N. This lies well outside the ^{15}N chemical shift range for imines (δ 305 to 365)[13] but within the ^{15}N chemical shift range for enaminoketones (δ 80 to 125). From these findings it is concluded that, in methanol solution used for recording the NMR spectra, the product exists >95% in the enaminoketone form (**17**). The enimine and ketimine forms were not detected. Previous investigations into the keto-enol tautomerism of Schiff bases prepared from monoamines and aliphatic dicarbonyl compounds have indicated that only on isomer, the enaminoketone, is present in solution. Dudek and Holm[14] have ascribed the preferential existence of an enaminoketone form, over that of the enamine and ketimine, to greater stabilisation through hydrogen bonding.



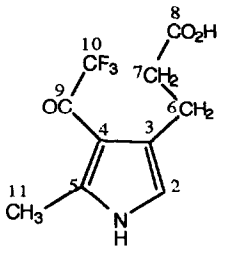
(18)

Although it could not be detected directly, the existence of the enol form of enaminoketone (**18**), in rapid exchange with the keto form (**17**), was indirectly demonstrated by monitoring hydrogen exchange at the C_6 methylene group of (**17**) 50% enriched in ^{15}N by ^{1}H NMR spectroscopy in deuteriated methanol. The 6-CHD proton of (**17**) appeared as a 1:1:1 triplet at δ4.50 while its ^{15}N-H resonance appeared as a doublet due to coupling with the proton of the α-CHD group attached to the nitrogen atom. The ^{15}N-H resonance of the dideuteriated form of **17** appeared as a doublet, one on either side of its ^{14}N-H resonance at δ11.3. The formation of (**18**) is important in considering the mechanism of the subsequent cyclisation to form a pyrrole, a matter to be considered in a subsequent publication.

If the reaction is carried out at reflux temperature cyclisation occurs to give the Knorr product 3-(2-carboxyethyl)-5-methyl-4-trifluoro-acetylpyrrole (**19**). This was inferred from the NMR data, particularly a signal with a chemical shift of δ 6.53, corresponding to α-CH of a pyrrole ring. There was no evidence for formation of the Fischer-Fink product. **19** was obtained when **17** was refluxed in acetate buffer and it is concluded that **17** is an intermediate in the formation of **19**.

Reaction of 5-ALA with 1,1,1,5,5,5,-hexafluoropentane-1,2-dione There appears to be no reaction between these two substances. Some 2,5-bis(2-carboxyethyl)dihydropyrazine, formed by the dimerisation of 2 molecules of 5-ALA, was detected but the dione was recovered unreacted. It exists in aqueous solution as a dihydrate and this is clearly an unreactive form .

Reaction of 5-methyl-5ALA with pentane-2,4-dione. Analysis of the solid product by NMR spectroscopy revealed that the only product formed was the Knorr product, 3-acetyl-4-(2-carboxyethyl)-2,5-dimethylpyrrole (**20**). Formation of the Fischer-Fink pyrrolic product is not possible unless methanol is eliminated. This appears not to happen.



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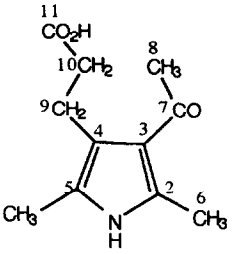

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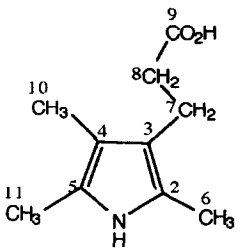


(21)

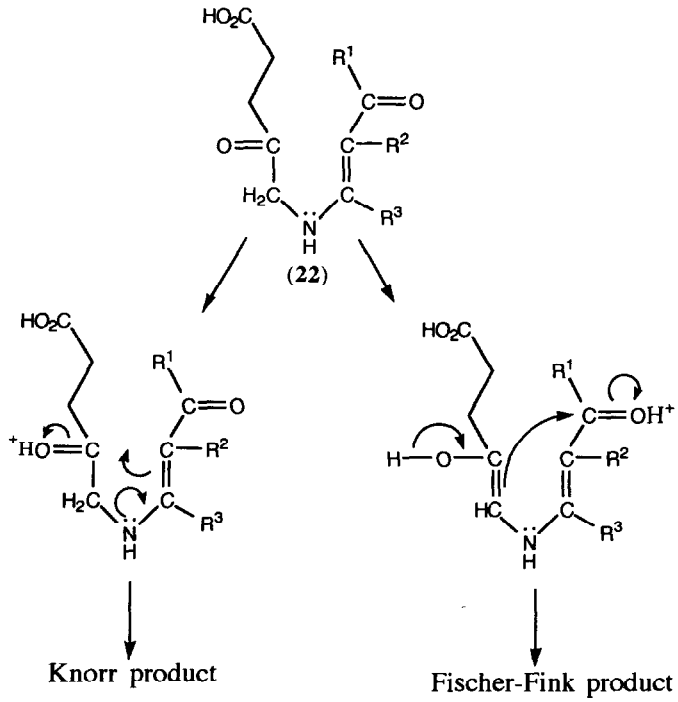
Reaction of 5-methyl-5-ALA with 3-methylpentane-2,4-dione. During the time of reflux used for the other reactions in this series no solid product was obtained. However, examination of the solution by ^{13}C NMR spectroscopy revealed formation of 2,5-bis(2-carboxyethyl)-3,6-dimethylpyrazine, as well as unreacted reactants. Prolonged reaction lead to the production of a very small amount of 3-(2-carboxyethyl)-2,4,5-trimethylpyrrole (**21**) formed by hydrolysis of the Knorr product. Formation of a Fischer-Fink product is not possible.

DISCUSSION

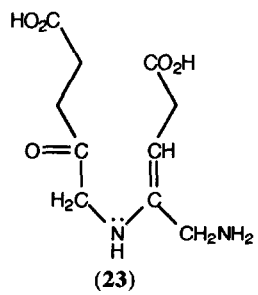
From our observation that 1,1,1,5,5,5-hexafluoro-pentane-2,4-dione, which exists exclusively in the keto form or its hydrate, does not react at all with 5-ALA we conclude that the reactive form of the dione is most likely to be an enol. The form of 5-ALA in aqueous which reacts with the dione cannot be determined from the results reported as all the forms reported by Jaffe and Rajagopalan[9] have an amino group nucleophilic enough to react with the enolic form of the dione. It is reasonable to assume that the intermediate formed as the first product of reaction is the enaminoketone **22** This is inferred from the isolation and identification of **18** from the reaction of 5-ALA with 1,1,1-trifluoropentane-2,4-dione and by the fact that no pyrrolic products were obtained from reaction with 3,3-dimethylpentane-2,4-dione. If **22** can exist in as many forms as does 5ALA then two enolic forms of **22** are possible intermediates in the subsequent cyclisation step. Indeed, one them, the dominant one according to the observations of Jaffe and Rajagopalan[9], will be used to explain formation of the Fischer-Fink product. Although it seems possible that **22** forms a hydrate the cyclisation process can be explained without evoking this species.

Proposed routes from 22 to the cyclised products are shown in Scheme 3. Protonation of the 4-keto-group of the 5-ALA side of **22** leads readily to cyclisation and elimination of water, in the manner shown in the Scheme, to give the Knorr product. Enolisation of that keto group and protonation of the other keto-group leads, in the manner shown in the Scheme, to the Fischer-Fink product We know that enolisation at the C-5 position does occur in 5-ALA and should occur in a similar way in **22** but this is not the only factor which decides whether cyclisation is Knorr or Fischer-Fink. We have also to consider the relative ease of protonation of the two keto groups and, with so many factors it is not surprising therefore, that in the cases of pentane-2,4-dione and hexane-2,4-dione both pyrrolic products are obtained. When R^1 is the electron withdrawing group CF_3 protonation of the neighbouring keto-group will be disfavoured and only the Knorr product is obtained. From an examination of the Taft substituent constants the effect of an alkyl group in a aliphatic system is seen to be electron donating and so the keto group in 3-methyl and 3-propylpentane-2,4-dione will be more basic and so the Fischer-Fink product is obtained as factors leading to the Knorr product will be unaffected by this substitution. The polar effect of an ethoxy group is electron withdrawing and so with ethyl acetoacetate only the Knorr product is obtained. Substitution of a methyl group at the 5-position in 5-ALA disfavours enol formation in **22** and so only the Knorr product is obtained.

The equivalent of **22** formed from two molecules of 5-ALA i e. **23** is suitable for cyclisation in the Knorr sense and it might be imagined that PBG would be formed in a nonenzymatic reaction but this appears not to be the case. However it is clear that **23** could be formed only from the C_3 enol of 5-ALA and we know that this is disfavoured with respect to the C_5 enol. It seems possible that the observation by Jaffe and Rajagopalan[9] that there is preferred enolisation to C_5 may explain the absence of spontaneous cyclisation to PBG. We await further elucidation of the mechanism employed by the enzyme to effect this step.



SCHEME 3



Along with the solid pyrrole products obtained in the manner described above, the filtrate obtained from the reaction mixture contained some 2,5-(2-carboxyethyl)dihydropyrazine. This product of reaction was identified by Granick and Mauzerall[7] and its ^{13}C NMR spectrum reported by Jaffe and Rajagopalan[9]. They obtained this compound by the spontaneous cyclisation of 5-ALA in acid buffer. Its mode of formation has been commented upon[10]. In the presence of air oxidation to a pyrazine can occur and when the substrate was 5-methyl-5-ALA the product observed by ^{13}C NMR spectroscopy was 2,5-bis(2-carboxyethyl)-3,6-dimethylpyrazine.

EXPERIMENTAL

Materials. 5-ALA.HCL was purchased from the Sigma Chemical Company. All other chemicals were AnalaR grade whereever possible. Carbon-13 labelled 5-ALA at the 4-position was made by minor modifications of the methods of Pichat and Herbert[15] and Tschudy and Collins.[16] Nitrogen-15 labelled 5-ALA was made from labelled potassium phthalimide and methyl 5-chlorolevulinate by the method of Neuberger and Scott.[17] 5-methyl-ALA.HCl was made by the reaction of potassium phethalimide and methyl 5-chloro-5-methyllevulinate by the method of Lartillot and Baron.[18] 3-Methylpentane-2,4-dione and 3,3-dimethylpentane-2,4-dione were prepared by the method of Mao et al.[19]

Techniques. NMR spectra were obtained on a Bruker AM300 spectrophotometer with DSS as a standard for aqueous solutions. For the nitrogen spectra the standard was external nitromethane. All spectra were acquired at a probe temperature of 37°C. A Griffin digital pH meter was used.

Reaction of 5-ALA with pentane-2,4-dione. The following is a typical experimental procedure for this study A solution of 5-ALA HCl (0.0838 g; 0.5 mmol) and pentane-2,4-dione (0 05 g; 0.5 mmol) in acetate buffer (2ml; pH = 4.6) was heated under reflux for 0.5h. The reaction mixture was cooled and the product filtered off, washed thoroughly with cold water, and dried over P_2O_5 *in vacuo* to constant weight (0.0605 g; 62%), m.p. 191°C. (Found: C, 61 5, H, 6.7; N, 7.1. $C_{10}H_{13}NO_3$ requires C, 61.5, H, 6.7; N, 7.2%). NMR spectra allowed us to identify the two products: 3-acetyl-4-(2-carboxyethyl)-2-methylpyrrole(**8**) [$\delta_C(CD_3OD)$ 138.0 (C_2), 121.0(C_3), 125.8 (C_4), 116.2 (C_5), 15 4 (C_6), 197.6 (C_7), 30.7 (C_8), 24.2 (C_9), 35.9 (C_{10}), 177 6 (C_{11}); δ_H (CD_3OD) 6.40 (s, 5-CH), 2.47 (s, 6-CH_3), 2 4 (s, 8-CH_3), 2.94 (t, 9-CH_2, J = 7.42 Hz), 2.53 (t, 10-CH_2, J = 7.42 Hz)] and 2-(3-carboxypropanoyl)-3,5-dimethylpyrrole(**9**) [$\delta_C(CD_3OD)$ 129.0(C_2), 131.0 (C_3), 113.4 (C_4), 136.4 (C_5), 189.2 (C_6), 35.0 (C_7), 27.1 (C_8), 176.9 (C_9), 12.8 (C_{10}), and 14.6 (C_{11}), $\delta_H(CD_3OD)$ 5.80 (s, 4-CH), 3.01 (t, 7-CH_2, J = 6.60 Hz), 2.64 (t, 8-CH_2, J = 6.60 Hz), 2.21 (s, 10-CH_3) and 2 32 (s, 11-CH_3).

Reaction of 5-ALA with ethyl acetoacetate. Only one product was obtained (59%), m.p. 163°C. (Found. C,59.0; H, 6 7; N, 6.2 $C_{11}H_{14}NO_4$ requires C, 58.7; H, 6.7; N, 6.2%). The NMR spectra showed the product to be 3-(2-carboxyethyl)-4-ethoxycarbonyl-5-methylpyrrole (**10**) [$\delta_C(CD_3OD)$ 115.7 (C_2), 125 2 (C_3), 110 1 (C_4), 138 (C_5), 23.7 (C_6), 36 4 (C_7), 176 0 (C_8), 168 0 (C_9), 60.2 (C_{10}), 14.8 and 13.9 (C_{11} and C_{12}) $\delta_H(CD_3OD)$ 6.36 (s, 2-CH), 2.92 (t, 6-CH_2, J = 7 70 Hz), 2 56 (t, 7-CH_2, 7.70 Hz), 4.22 (q, 10-CH_2, J =7.15 Hz), 1.33 (t, 11-CH_3, J = 7.15 H_2), 2 41 (s, 12-CH_3)]

Reaction of 5-ALA with 3-methyl pentane-2,4-dione. Only one product was obtained (16.7%) m p. 218°C (Found. C, 63.2; H, 7.2; N, 6.7 $C_{11}H_{15}NO_3$ requires C, 63 1, H, 6.7%). The NMR spectra showed the product to be 2-(3-carboxypropanoyl)-3,4,5-trimethylpyrrole(**11**) [$\delta_C((CH_3)_2SO)$ 125.6, 126.2, 116 4, 131.1 (C_2-C_5),186 4 (C_6), 33.5 (C_7), 27.8 (C_8), 174.0 (C_9), 10 9, 8.4, 11.3 (C_{10}-C_{12}) $\delta_H((CH_3)_2SO)$ 2.90 (t,7-CH_2, J = 6.60 H_2), 2.50 (t, 8-CH_2, J = 6 60H_2), 2 13, 1 85, and 2.19 (s, 10-CH_3-12CH_3)]

Reaction of 5-ALA with 3-isopropylpentane-2,4-dione Only one product was obtained (10%) m.p. 178°C (Found: C, 65 8; H, 8 0, N, 5 8 $C_{13}H_{19}NO_3$ requires C, 65.8; H, 8.1; N,5 9%) The NMR spectra showed the product to be 2-(3-carboxypropanoyl)-3,5-dimethyl-4-isopropylpyrrole(**12**) [$\delta_C((CD_3)_2SO)$ 126.0, 126.5, 124.7 and 129.7 (C_2-C_5), 189.7 (C_6), 34 7 (C_7), 29.8 (C_8), 174.8 (C_9) 11.4 and 12.2 (C_{10} and C_{13}), 24.3 (C_{11}), 22.4 (C_{12}) $\delta_H((CD_3)_2SO)$ 2.84 (t, 7-CH_2 J = 6.95 Hz), 2 39 (t, 8-CH_2, J = 6.95 Hz), 2.20 and 2.25 (s, 10-CH_3 and 13-CH_3), 2 87 (m, 11-CH, J = 6.95 Hz), 1 18 (d, 12-CH_3, J = 6 95)].

Reaction of 5-ALA with hexane-2,4-dione. Gas chromatography-mass spectrometry showed that the product of reaction (59 7%) consisted of 4 components, all with molecular ion peaks of the same relative molecular mass (m/z = 209), corresponding to the four modes of cyclisation. The NMR spectra were consistent with a mixture of 3-(2-carboxyethyl)-5-methyl-4-propanoylpyrrole (**13**) [$\delta_C(CD_3OD)$ 116.1 (C_2), 125.7 (C_3), 120 6 (C_4), 116.1 (C_5), 24 3 (C_6), 35.9 (C_7), 177 6 (C_8), 200 8 (C_9), 36 0 (C_{10}), 9 0 (C_{11}), 15.5 (C_{12}) δ_H (CD_3OD), 6 40 (s, 2-CH), 2 95 (t, 6-CH_2, J = 7 29 Hz), 2 54 (t, 7-CH_2, J = 7.53 Hz), 2 75 (q, 10-CH_2, J = 7.53 Hz), 1.13 (t, 11-CH_3, J = 7 29 Hz), 2 47 (s, 12-CH_3)], 3-acetyl-4-(2 carboxyethyl)-2-ethylpyrrole(**15**) [143.8 (C_2),

120.1 (C_3), 125.4 (C_4), 116.5 (C_5), 22.8 (C_6), 14.2 (C_7), 197.8 (C_8), 30.5 (C_9), 24.2 (C_{10}), 35.8(C_{11}), 177.5 (C_{12}) δ_H (CD_3OD) 6.43 (s, 5-CH), 2.89 (q, 6-CH_2, J = 7.53 Hz), 1.23 (t, 7-CH_3, J = 7.29 Hz), 2.42 (s, 9-CH_3), 2.95 (t, 10-CH_2, J=7.29 Hz) 2.55 (t, 11-CH_2, J=7.53 Hz)], 2-(3-carboxypropanoyl)-3-ethyl-5-methylpyrrole(**14**) [δ_C (CD_3OD) 116.2 (C_2), 136.5 (C_3), 111.2 (C_4), 137.3 (C_5), 189.1 (C_6), 34.7 (C_7), 29.2 (C_8), 176.9 (C_9), 66.9 (C_{10}), 15.1 and 12.9 (C_{11} and C_{12}) δ_H (CD_2OD) 5.87 (s, 4-CH), 3.02 (t, 7-CH_2, J = 6.60 Hz), 2.65 (t, 8-CH_2, J = 6.60 Hz), 3.48 (q, 10-CH_2, J = 7.29 Hz) 1.17 (t, 11-CH_3, J = 7.29 Hz), 2.23 (s, 12-CH_3)], 2-(3-carboxypropanoyl)-3-methyl-5-ethylpyrrole(**16**) [δ_C (CD_3OD) 116.5 (C_2), 138.3 (C_3), 111.8 (C_4), 144.6 (C_5), 189.3 (C_6), 35.0 (C_7), 29.2 (C_8), 176.0 (C_9), 14.7 and 14.0 (C_{10} and C_{12}), 22.2 (C_{11}) δ_H (CD_3OD) 5.84 (s, 4-CH), 3.02 (t, 7-CH_2 J = 6.60 Hz), 2.65 (t, 8-CH_2, J = 6.60 Hz), 2.33 (s, 10-CH_3), 2.76 (q, 11-CH_2, J = 7.29 Hz), 1.19 (t, 12-CH_3, J = 7.53)

Reactions of 5-ALA with 1,1,1-trifluoropentane-2,4-dione. The reaction mixture was allowed to stand overnight at room temperature and the fine, white, crystalline precipitate filtered off. After drying *in vacuo* the enaminoketone (**17**) was examined and its identity verified (Found: C, 45.0; H, 4.2; N, 5.1. $C_{10}H_{12}NO_4F_3$ requires C, 45.0; H, 4.5; N, 5.2%) [δ_C(CD_3OD) 125.0, 121.2, 117.4, 113.6 (C_1, $^1J_{CF}$ = 287.6 Hz), 176.8, 176.0, 175.6 (C_2, $^2J_{CF}$ = 32.3 Hz), 90.3 (C_3), 172.4 (C_4), 20.0 (C_5), 53.7 (C_6), 204.1 (C_7), 35.3 (C_8), 28.7 (C_9), 176.1 (C_{10}) δ_H (CD_3OD) 5.43 (s, 3-CH), 2.10 (s, 5-CH_3) 11.3 (s, NH), 4.52 (s, 6-CH_2), 2.77 (t, 8-CH_2, J = 6.36 Hz), 2.65 (t, 9-CH_2, J = 6.36 Hz)].

Reaction of 5-methyl-5-ALA with pentane-2,4-dione. Only one product was formed (16%) m.p. 222-224°C (Found: C, 61.7; H, 7.6; N, 6.4. $C_{11}H_{15}NO_3$ required C, 62.1; H, 7.7 N, 6.7%). The NMR spectra showed the product to be 3-acetyl-4-(2-carboxyethyl)2,5-dimethylpyrrole (**20**) [δ_C((CD_3)$_2$SO) 133.1 (C_2 119.7 (C_3), 122.8 (C_4), 118.3 (C_5), 14.8 (C_6) 193.3 (C_7), 30.4 (C_8), 20.8 (C_9), 35.1 (C_{10}), 174.5 (C_{11}), 9.7 (C_{12}) δ_H((CD_3)$_2$SO) 2.38 (s, 6-CH_3), 2.27 (s, 8-CH_3), 2.72 (t, 9-CH_2, J = 7.74 Hz), 2 29 (t, 10-CH_2, J = 7.74 Hz), 2.03 (s, 12-CH_3)].

Reaction of 5-methyl-5-ALA with 3-methylpentane 2,4-dione. The identification of a pyrazine has been described previously. The pyrrole (**21**) was identified from its NMR spectra (δ_C(CD_3OD) 122.4, 122.1 (C_2 and C_5), 116.6 (C_3, 112.6 (C_4) 10.8 and 10 7 (C_6 and C_{11}), 21.1 (C_7), 37.0 (C_8), 179.2 (C_9, 9.1 (C_{10}) δ_H (CD_3OD) 2.09, 2 10 (s, 6-CH_3, 11-CH_3), 2.63 (t, 7-CH_2, J = 8.25 Hz) 2.38 (t, 8-CH_2, J = 8.25 Hz), 1.88 (s, 10-CH_3)

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