CHAPTER II

HISTORY

Valproic acid (X), [VPA ; di-n-propylacetic acid , DPA;2-propylpentanoic acid, or 2-propylvaleric acid].

The acid,a colourless liquid, was first synthesized by Burton (Burton, 1882, quoted in Chapman et al.,1982). No therapeutic application of the drug was known until the discovery of its anticonvulsant properties in February, 1962 by P.Eymard in the laboratories of the Berthier firm in Grenoble. The conditions of this discovery were described at a meeting of the French Society of Therapeutics and Pharmacodynamics on December 19, 1962, and subsequently published in the journal Therapie (1963 , XVIII, 435-438): "Having synthesized a series of derivatives of khelline, P.Eymard started a study of their pharmacological properties in the laboratory of G.Carraz, but was impeded by their solubility in water and the usual organic solvents. H.Meunier, Y.Meunier and P.Eymard in the research department of Berthier Laboratories had the idea of dissolving the most active molecule in n-dipropyl acetic acid , which H. and Y. Meunier used regularly for the preparation of a bismuth salt. This solvent revealed certain pharmacological properties..". G. Carraz requested

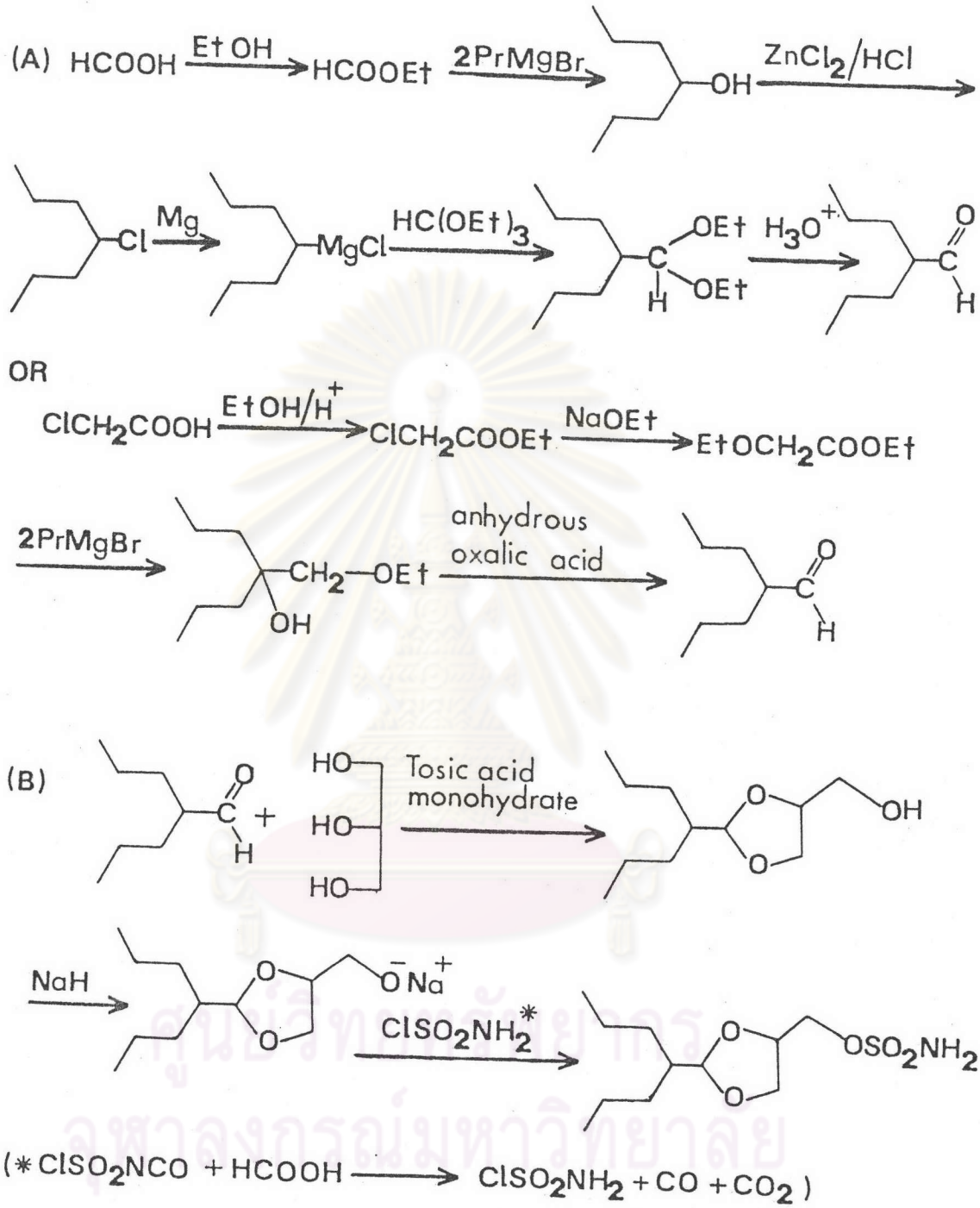


Figure 11. The synthetic approach of [2-(1-propylbutyl)-1,3-dioxolan-4-yl]methyl sulfamate

that it be studied in a wide battery of tests, including the properties against pentylenetetrazol induced seizures. The test solution exhibited a marked protection in this test. Subsequent studies indicated that the anticonvulsant action was due to the solvent, valproic acid. Results from many studies show that valproic acid has a very broad spectrum of anticonvulsant activity, probably greater than that of any other anticonvulsant in clinical use (Chapman et al., 1982).

Clinical efficacy of valproic acid

Valproic acid's broad spectrum of an anticonvulsant activity in animal seizure models is reflected in its diverse clinical utility. Although its original indication was for the treatment of absence seizures, valproic acid also appears to be effective against certain myoclonic seizures , generalized tonic-clonic seizures, and perhaps partial seizures (Rogawski and Porter, 1990).

Metabolism

Valproic acid is a branch-chain fatty acid with an uneven number of carbons on each chain. The metabolism of valproate appears to be complex. Valproate is almost completely metabolised before excretion, only 1-3% of the administered dose being found as unchanged drug in the

urine. Four metabolic pathways have been found for valproate: glucuronidation, β-oxidation, ω-oxidation, and ω-1 oxidation (Figure 12) (Gugler and von Unruh, 1980).

In the dose range normally used glucuronidation is the main metabolic route for valproate. The acute anticonvulsant activity of metabolites of valproate has been evaluated against electroshock and pentylenetetrazol seizures in mice. None is as potent as valproic acid. The most active compound is 2-propyl-4-pentenoic acid(Loscher, 1981).

Analogues:Structure and anticonvulsant action

Since the discovery of the therapeutic properties of valproate many analogues have been tested for their anticonvulsant action. The ability to protect against pentylenetetrazol induced mortality in mice is shown for four classes of valproate analogues in Figure 13. Among the closest analogues of valproate, the anticonvulsant activity is maintained when the acids contain 6-8 carbon atoms. Branching or unsaturation of one or both chain does not result in the loss of activity. Analogues in which the carboxylic group is separated from the branching of the chain by one or more carbons, are anticonvulsant but with a marked sedation action. Cyclic analogues of valproate have also been studied. Methyl-1-cyclohexane

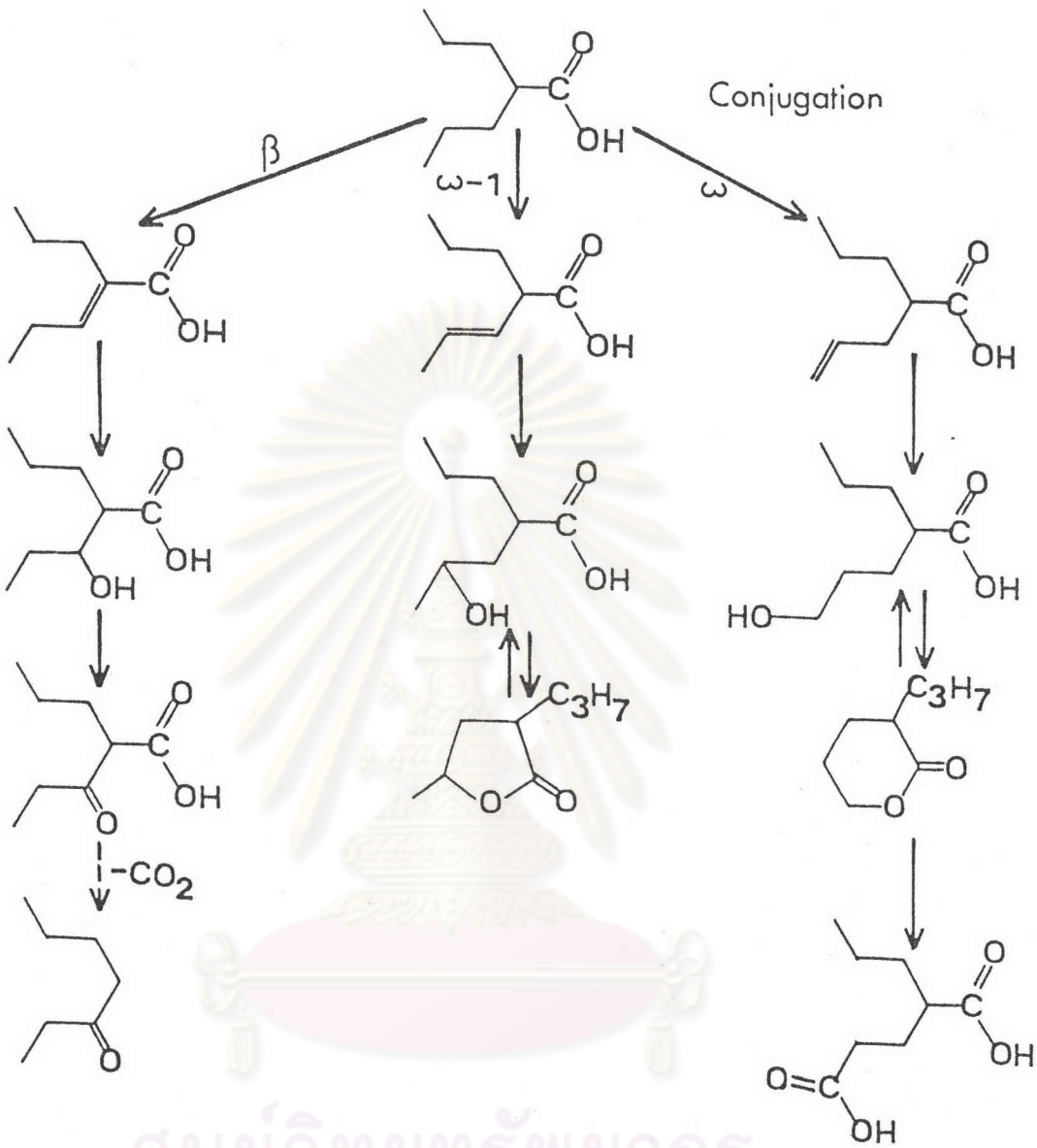


Figure 12. Metabolic pathway of valproate

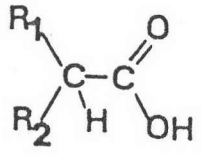
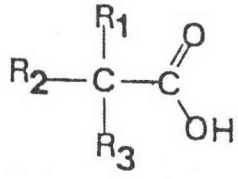
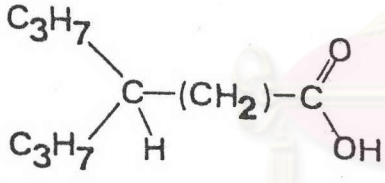
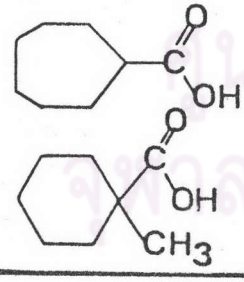
STRUCTURE				ACTIVITY*
(I)		R_1	R_2	
		C_3H_7	C_3H_7	100
		C_2H_5	C_2H_5	80
		C_4H_9	C_4H_9	0
		C_2H_5	C_4H_9	100
		$(CH_3)_2$ CH	C_3H_7	60
		$CH_2{=}CHCH_2$	$CH_2{=}CHCH_2$	100
		$HC{\equiv}CCH_2$	$HC{\equiv}CCH_2$	50
(II)		R_1	R_2	R_3
		CH_3	C_3H_7	C_2H_5 100
		CH_3	C_4H_9	C_2H_5 100
		CH_3	C_3H_7	CH_3 50
		C_3H_7	C_3H_7	C_3H_7 100
(III)			n	
			1	100
			2	100
			3	50
(IV)				40
				100

Figure 13. Anticonvulsant activity of analogues of valproate

(*% protection against mortalily produced by pentylenetetrazol. Drugs were administered to mice at the dose of 1.39 mmol/kg, ip., 30 min prior to pentylenetetrazol.)

carboxylic acid is as active as valproate.

The, amide and various esters of valproate have also been found to be anticonvulsant, Dipropylacetamide appears to owe its anticonvulsant activity in man to its conversion to valproate in the gastrointestinal tract. The amide also possesses intrinsic anticonvulsant activity(Chapman et al., 1982).

Mechanisms of action

Two general hypotheses have been proposed to explained the antiepileptic activity of valproate. The first of these proposes that valproate acts by enhancing GABA - mediated inhibition and relies primarily on data demonstrating that the drug increases brain GABA levels. The second hypothesis posits a phenytoin -like effect of valproate on voltage-dependent Na^+ channel.

1. Effect on GABA system

There is an increase in plasma and CSF-levels of GABA in patients being treated with usual clinically effective doses of valproate which is compatible with the idea that effects on GABA matabolism may be relevant to the antiepileptic activity of the drug in human subjects. However the mechanism by which valproate increases GABA

levels is not well understood. The drug inhibits several enzymes involved in GABA degradation, including GABA-T, succinic semialdehyde dehydrogenase (SSADH), and aldehyde reductase (ALD.Rase). In addition, valproate may also increase the activity of glutamic acid decarboxylase (GAD), the enzyme responsible for GABA synthesis. Figure 14 shows the metabolism of L-glutamate.

Since SSADH is much more sensitive to valproate inhibition than GABA-T, it has been argued that the main mechanism for the valproate-induced increase in brain GABA levels is the primary inhibition of SSADH by valproate with a resulting accumulation of SSA , which in turn inhibits the forward GABA-T reaction , leading to an increase GABA level (Chapman et al., 1982 ; Nanavati and Silverman, 1989; Rogawski and Porter, 1991).

2. Block of voltage-dependent Na^+ channels.

Like phenytoin and carbamazepine, valproate limits the ability of cultured CNS neurous to fire Na^+-dependent action potentials at high frequency. The use- and voltage-dependent effects of valproate on Na^+ conductance are compatible with the requirements during a seizure while having a minimal effect on normal neuronal firing.

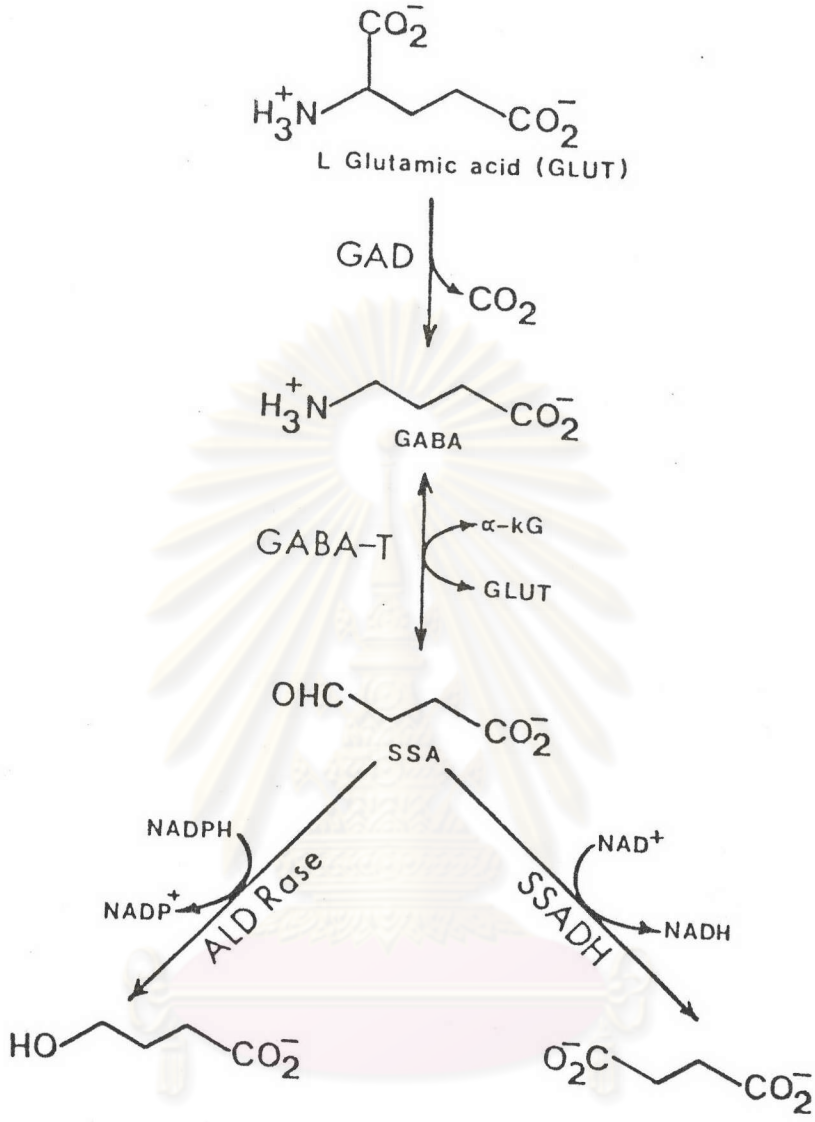


Figure 14. Metabolism of GABA

Because valproate has such a wide spectrum of anticonvulsant activity, it is attractive to accept the view that the drug's clinical activity may relate to a combination of mechanisms. Nevertheless,these observations highlight the fact that full spectrum of pharmacological actions of valproate are not yet well understood (Rogawski and Porter, 1991).

I. Synthesis of aldehyde : 2-propylpentanal.

Since the formation of 2-alkyl-1,3-dioxolan-4-methanol can be obtained from the condensation of corresponding aldehydes and glycerol, valproic acid must then be prepared as 2-propylpentanal so that it can react with glycerol to form [2-(1-propylbutyl)-1,3-dioxolan-4-yl] methanol. Aldehyde can be synthesized from various types of reactions as described below.

A. Syntheses of aldehydes from Grignard reagents (RMgX).

A.1 Reactions of ethyl ethoxyacetate ($EtOCH_2COOEt$) with Grignard reagents (Figure 15-A)

Behal and Sommelet (1904) prepared aldehydes of the type R_2CHCHO from Grignard reagents, RMgX by the use of ethyl ethoxyacetate. The alcohols,$EtOCH_2(OH)R_2$

formed were then transformed to aldehydes by the use of acid. The yields varied between 50-80% of the theoretical quantity.

A.2 Reactions of ethyl orthoformate [$CH(OEt)_3$] with Grignard reagents. (Figure 15-B)

This method is also known as Bodroux-Tschitschibabin aldehyde synthesis since Bodroux and Tschitschibabin independently discovered it in 1904. They discovered that if ethyl orthoformate was first added to the ethereal solution of the Grignard reagent, followed by refluxing the reaction mixture for several hours, and then most of the solvent was removed by distillation, a point was reached at which a vigorous reaction ensued. After that the reaction product , acetal was refluxed with acid in order to be decomposed to aldehyde (Smith and Bayliss, 1941; Smith and Nichols, 1941).

A.3 Reactions of N,N-disubstituted formamide (R'R"NCHO) with Grignard reagents (Figure.15-C)

This method is also known as Bouveault aldehyde synthesis since Bouveault discovered it in 1903. The equimolecular amount of formamide was added to the ethereal solution of the Grignard reagent. Then the reaction product was decomposed to aldehyde by acid.

A. $EtOCH_2COOEt + 2RMgX \longrightarrow EtOCH_2C(OH)R_2$

$EtOCH_2C(OH)R_2 \xrightarrow{H^+} EtOCH{=}CR_2 \longrightarrow HOCH{=}CR_2$

$\longrightarrow R_2CHCHO$

B. $CH(OEt)_3 + RMgX \longrightarrow RCH(OEt)_2 + MgXOEt$

$RCH(OEt)_2 \xrightarrow{H_3O^+} RCHO + 2EtOH$

C. $RMgX + R'R''NCHO \longrightarrow RCH(OMgX)NR'R''$

$\xrightarrow{H_3O^+} RCHO + R'R''NH + MgX_2$

D. $RMgX + CS_2 \longrightarrow RCSSH$

$RCSSH + H_2NCONHNH_2 \longrightarrow RCH{=}NNHCONH_2 + H_2S + S$

$RCH{=}NNHCONH_2 + HCl + H_2O \longrightarrow RCHO + H_2NCONHNH_2.HCl$

Figure 15. Synthesis of aldehydes from Grignard reagents and

A. ethyl ethoxyacetate;

B. triethyl orthoformate;

C. N,N-disubstituted formamide;

D. Carbon disulfide

(Smith and Bayliss, 1941; Smith and Nichols, 1941).

, A.4 Reactions of carbon disulfide (CS_2) with Grignard reagents. (Figure 15-D)

Wuyts and his collaborators discovered and developed a synthesis of aldehyde in which a Grignard reagent reacted with carbon disulfide to produce a dithio acid which was in turn, converted into an aldehyde derivative by action of semicarbazide, phenylhydrazine or hydroxylamine. The aldehyde derivative was then hydrolyzed to the aldehyde (Smith and Nichols, 1941).

B. The oxidation of primary alcohols to aldehydes (Figure 16-A)

Simple aldehydes may be obtained in reasonably good yield by oxidation of the corresponding primary alcohols with sodium dichromate in dilute sulfuric acid solution. To avoid further oxidation to the corresponding carboxylic acids, the aldehydes are removed as rapidly as possible by distillation through a fractionating column.

These rather vigorous conditions (high temperature and/or an aqueous strongly acidic enviroment) are however unsuitable for those primary alcohols which are insoluble in water. For those reasons several

non-aqueous chromium (VI) oxidising reagents have been developed including the Collins reagent , pyridium-chlorochromate and pyridium dichromate.

Corey and Schmidt(1979); Furniss et al. (1991) had investigated that the reagent pyridium dichromate in methylene chloride oxidized primary alcohols to the corresponding aldehydes and no further, regardless of the nature of the substrate.

C. The reduction of nitriles (RCN) to aldehydes (Figure 16-B)

The reduction of a nitrile is achieved with anhydrous tin (II) chloride dissolved in ether or ethyl acetate saturated with dry hydrogen chloride (the Stephen aldehyde synthesis). The resulting aldimine hydrochloride (probably in the form of a complex with tin (IV) chloride) is then hydrolyzed in warm water.

The method is used mainly for the synthesis of aromatic aldehydes but reduction of the higher aliphatic nitriles normally gives good yields (Stephen, 1925).

D. The reduction of acid chlorides (ROCl) to aldehydes (Figure 16-C)

Brown and McFarlin(1958) had investigated that lithium tri-t-butoxyaluminohydride obtained from the reaction of lithium aluminium hydride and t-butyl alcohol was a selective reducing agent. They observed that when lithium tri-t-butoxyaluminohydride in tetrahydrofuran or diglyme solution added in stoichiometric amount to the acid chloride in the same solvent at -80°C, gave the aldehyde in yields of 60 to 80% in the case of aromatic derivatives and 40 to 60% in the case of aliphatic derivatives.

E. The reduction of carboxylic acids to aldehydes (Figure 16-D)

Bedenbaugh et al. (1970) discovered that corboxylic acids were reduced to aldehydes by lithium in methylamine. One reduction intermediate is thought to be a carbinolamine salt which is converted to the imine during the isolation procedure. The imine (which can be isolated in good yield) is hydrolyzed to the aldehyde (slowly in neutral aqueous solution, rapidly in acidic aqueous solution). They investigated that saturated acids ranging from five to fourteen carbons, an unsaturated acid (8-octadecenoic), and a diacid (nonadioic) were reduced with good results (yield ~ 60%).

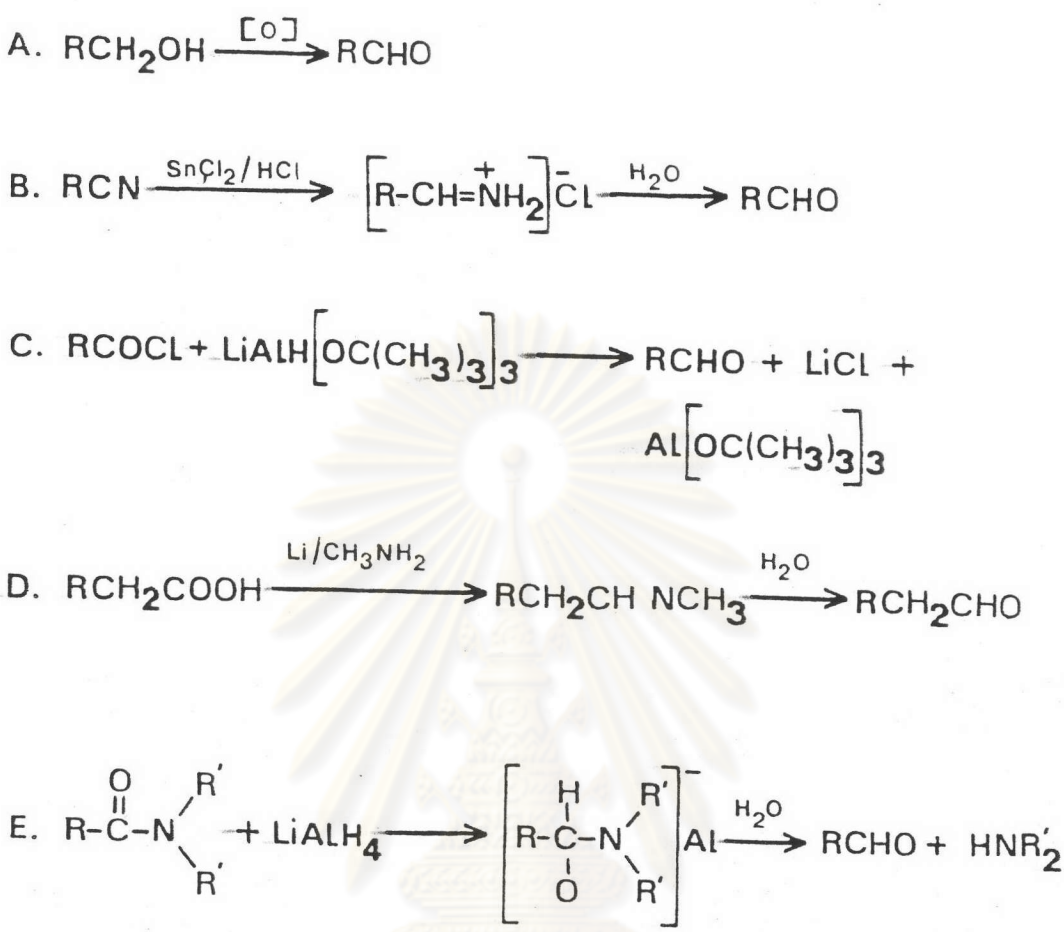


Figure 16. Synthesis of aldehydes from the oxidation of A. primary alcohols ; and the reduction of B. nitriles; C.acid chlorides ; D. carboxylic acids; E. acid amides

F. The reduction of acid amides to aldehydes. (Figure 16-E)

Micovic and Mihailovic (1953) discovered that if the ethereal solution of lithium aluminium hydride was added to the ethereal solution of amide at low temperature (-15°C to -10°C) and with 0.25 mole of hydride for one mole of disubstituted amide, aldehydes were obtained. The formation of aldehydes probably proceeds through the complex formed as the result of a usual nucleophilic substitution, under the attacks of the hydride in AlH^-_4. Upon hydrolysis, this complex would give an unstable amino alcohol which would decompose , by intermolecular displacement, to aldehyde and the starting amine.

II. Syntheses of 2-substituted-1,3-dioxolan-4-methanols.

Cyclic acetals can be prepared from condensation of glycerol and aldehyde in the presence of the acid catalyst such as p-toluenesulfonic acid monohydrate. The removal of water formed is found materially to improve the yield of acetal. This can be achieved either by azeotropic distillation or the use of dehydrating agents such as anhydrous calcium chloride. (Showler and Darley, 1967; Vicchio and Callery, 1989). (Figure 17-A)

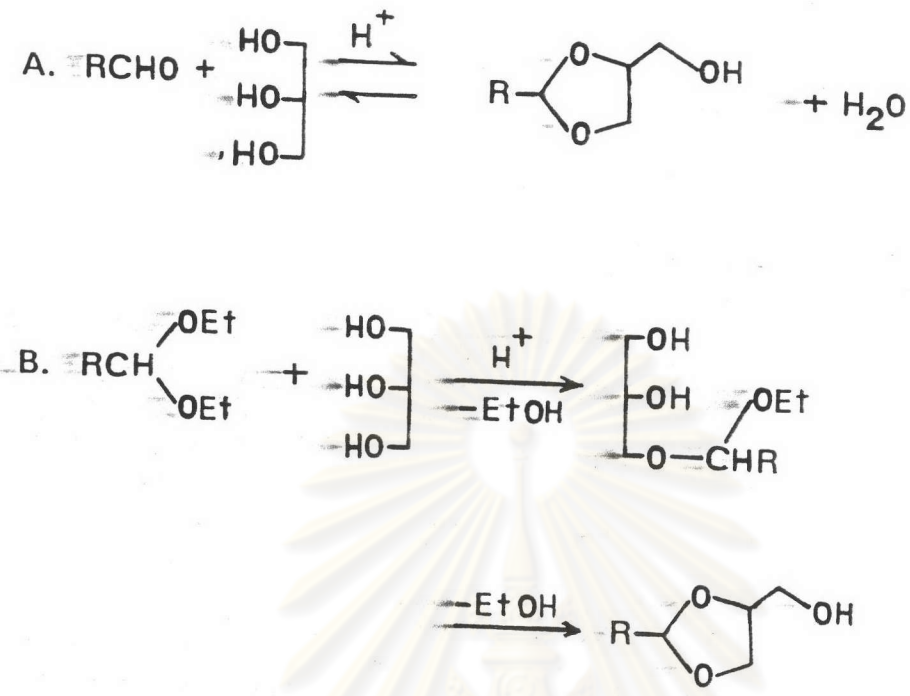


Figure 17. Synthesis of 2-substituted-1,3-dioxolan-4-methanols

Transacetalation reactions (Figure 17-B)give cyclic acetals when aliphatic acetals and glycerol are condensed in the presence of sulfosalicylic acid(Piantadosi et al., 1958)

III. Syntheses of sulfamate derivatives ($ROSO_2NH_2$)

A. The reactions of sulfamoyl chloride ($ClSO_2NH_2$) with sodium alkoxides or alcohols (Graf, 1968; Maryanoff et al., 1987; Lo et al., 1992). (Figure 18-A)

B. The reaction of phenylsulfamate with alcohol (Figure 18-B)

The sulfamoyl group from phenol is transferrd to alcohol at 90°C to 120°C. This reaction is rapid and irreversible (Lohaus, 1972; Lo et al.,1992).

C. The reaction of (benzyloxycarbonyl) sulfamoyl chloride with alcohol. (Figure 18-C)

The reaction of alcohol with N-chlorosulfonyl-urethane such as (benzyloxycarbonyl) sulfamoyl chloride yields ester of urethane-N-sulfonic acid, which is then submitted to catalytic hydrogenation to yield sulfamate (Graf, 1963, 1968; Lo et al., 1992).

A. $RO^{\ominus}Na^{\oplus} + ClSO_2NH_2 \longrightarrow ROSO_2NH_2 + NaCl$

$ROH + ClSO_2NH_2 \longrightarrow ROSO_2NH_2 + HCl$

B. $PhOSO_2NH_2 + ROH \xrightarrow{90-120^{\circ}C} ROSO_2NH_2 + PhOH$

C. $ClSO_2NHCO_2CH_2Ph + ROH \longrightarrow ROSO_2NHCO_2CH_2Ph + HCl$

$ROSO_2NHCO_2CH_2Ph \xrightarrow{H_2 / Pd-C} ROSO_2NH_2$

D. $ROH + SO_2Cl_2 \longrightarrow ROSO_2Cl \xrightarrow{NaN_3} ROSO_2N_3$

$ROH + Cl_2SO_2N_3 \longrightarrow ROSO_2N_3$

$ROSO_2N_3 \xrightarrow{Reduction} ROSO_2NH_2$

Figure 18. Synthesis of sulfamate derivatives from the reactions

A. alcohol and sulfamoyl chloride;

B. alcohol and phenylsulfamate;

C. alcohol and (benzyloxycarbonyl)sulfamoyl chloride;

D. the reduction of azidosulfate

D. The reduction of azidosulfates ($ROSO_2N_3$) to sulfamates (Figure 18-D)

Azidosulfate can be prepared by reaction of alcohol either with sulfuryl chloride (SO_2Cl_2) and sodium azide (NaN_3) or chlorosulfonyl azide ($Cl_2SO_2N_3$). The azidosulfate is then reduced to sulfamate. The reduction reaction can be accomplished either by the action of catalytic hydrogenation (H_2/Pd-C) , sodium borohydride($NaBH_4$) ,or copper in methanol (Cu/MeOH) (Matier and Comer, 1972;Hedayatullah and Guy, 1975, 1978;Maryanoff et al., 1987).