Due to the latest developments in chemistry much advancement has recently been made in the field of plant products and infinite variety of structures are now known to occur naturally. Many of the compounds previously thought to be too unstable have now been isolated and identified. A very large number of structurally related compounds has been discovered and the biogenetic concepts for their formation have come to light in various cases. Considerable progress has been made regarding biogenesis and biosynthesis of the natural constituents within the last two decades inspite of the fact that many such substances quite commonly possess wonderful complex structures. Very recently many biogenetic substances (e.g. isoprenoids, triterpenes and steroids etc.) have been isolated also from the soluble fractions of organic material of many sediments and from identified fossils of various geological ages. The Chart indicates the general areas of metabolism in living organisms and the probable routes of formation of the various classes of organic compounds.

Among the building units for formation of the natural products by far the most important is considered to be the acetic acid; the other major spring-boards are the aromatic amino-acids;
*The term 'Acetogenins' (genesis from acetate) has been proposed by Richards and Hendrickson\(^\text{10}\) intended to include the compounds biogenetically derivable by the acetate hypothesis (excluding terpenoids) such as flavonoids, quinones, coumarins, chromones, depsides, benzophenones, and other oxygenated compounds most of which incorporate at least one benzene ring.
tryptophan, phenylalanine, and tyrosine, and the aliphatic amino-
acids: ornithine and lysine. After addition of methionine as a 
methylating agent to the above the list is supposed to be vir-
tually complete.

The role of acetate in the biosynthesis of almost all 
groups of natural products has received wide attention especially 
within the last about twenty-five years. Many structures may be 
correlated to acetate units and the importance of acetate as an 
intermediate in a host of biochemical processes has long been 
known.

Most of the reactions generating the natural compounds 
are enzyme-catalysed, and it is generally accepted that such 
reactions are mechanistically (and stereochemically) reasonable 
once in terms of ordinary organic reaction theory. It has also 
been suggested that some of the reactions taking place in the 
formation of such products may not be enzyme-catalysed.

Two major groups of the natural products: A and B 
(Chart - I), "The Terpenoids and Steroids" and "The Acetogenins", 
form the part of the theoretical portion of this thesis since 
majority of the products isolated and identified during the 
course of the present work belong mostly to these classes.
TERPENOIDS

Terpenoids form one of the most important, widespread, and chemically interesting classes of natural products, and they have drawn considerable attention of chemists during the last few decades\textsuperscript{2,5,6,8-12}. They are found mostly in plants, and recently an increasing number of such substances has been shown to be responsible for the biological activity of various systems.

Terpenoids are generally built up of C\textsubscript{5} (isoprene-I) units. The "Isoprene-Rule" (Wallach) has been a useful guiding principle\textsuperscript{12} for structural studies of these compounds. It states that the isoprene units are joined in head-to-tail fashion; although some isoprenoids are irregularly built, and a few, which do not obey the Isoprene-Rule, are also known. It has now been accepted that carbon-carbon rearrangements may occur during their formation in nature, and the idea of 'Biogenetic Isoprene Rule' was advanced by Ruzicka\textsuperscript{12} stating that carbon skeleton of a terpenoid may be deduced from a postulated simple precursor.

According to the Isoprene-Rule carbon skeletons of most of the C\textsubscript{10} (monoterpenoids) and C\textsubscript{15} (sesquiterpenoids) compounds may be derived from geraniol (II) and farnesol (III) by coiling up these molecules in different manner. But, it is not certain whether the ionic or radical processes are involved during the postulated cyclisation which may be dependent on reaction mechanism and conformation of the precursor as well as the
intermediate. The cyclisation and rearrangements of terpenoids in vivo are considered to be concerted\textsuperscript{13}, but this may not be necessarily the case with the rearrangements taking place in vitro.

Even the somewhat peculiar looking carbon skeletons of such compounds as $\alpha$-pinene (IV), fenchol (V), $\alpha$-santalene (VI), cedrene (VII), caryophyllene (VIII), and patchouli alcohol (IX) etc. are derivable either from that of geraniol or farnesol. $\beta$-Pinene (X) has been photosynthesized from its supposed\textsuperscript{14a} precursor myroene (XI). Various transformations have been proposed by different workers\textsuperscript{14,15} in this field.
The biogenetic conversion of farnesol (III) to the various sesquiterpenoids, e.g. caryophyllène (VIII), pyrethrosin (XII), α-eudesmol (XIII), humulene (XIV), and elemol (XV) etc. has been satisfactorily interpreted to be occurring through a cation intermediate as shown below (III → XV).

Stereochemical implications in the biogenesis of sesquiterpenoids have been discussed by Hendrickson. The ideas of Ruzicka and Barton on the involvement of large rings, in order to incorporate stereochemical features, have been extended.
The biogenesis of zierone (XVI), helminthosporal (XVII), copaene (XVIII), and various other sesquiterpenoids have been described by Barton \( ^{17} \), Buchi \( ^{18a} \), de Mayo \( ^{18b} \) and numerous other scientists \( ^{19} \).

The diterpenoids (C20) may be derived, in principle, geranyl-geraniol (XIX) or related substances containing four C$_5$ units joined in the regular manner. Manool (XX) and sclareol (XXI) are easily obtained and further transformation leads to the resin acids (e.g. abietic acid). The majority of diterpenes are
cyclic with no C₃-OH group and with the 5:10 'normal' configuration at the A:B ring junction that implies the stereo-specific cyclisation as shown (XXII → XXIII). A multitude of hypothetical routes have been proposed, and often acid-catalysed cyclisations in vitro parallel the proposed schemes. Recently several new structures have been discovered and their biogenetic routes suggested. But, the biosynthetic processes leading from the isoprenoid skeleton to the spirodihydrofuran system are not yet very clear. Interesting structures are represented by the bitter principle columbin (XXIV) and the macrocyclic compound cembrene (XXV). The new podolactones C(XXVI) and D(XXVII) are the first known terpenes to contain the sulphoxide grouping.
A novel class of C\textsubscript{25} terpenoids known as 'Sesterterpenoids' has been recently discovered\textsuperscript{21}. They may be derived from geranyl farnesol\textsuperscript{22a} (XXVIII). Sesterterpenes\textsuperscript{21,22} have been isolated from insect protective waxes, from fungal and several other sources. They form phytotoxic principles of a number of Helminthosporium and Cochliobolus species. Recently\textsuperscript{22a} the acyclic C\textsubscript{25} alcohols: geranyl-farnesol and geranyl-nerolidol,
have also been obtained of the natural origin. Ophiobolin Α(XXIX),
gascardic acid (XXX), ceroplastol (XXXI), and moenocinol\(^{23a}\)(XXXII)
etc. are some of the members of this type (Chart - II).

Chilanthatriol\(^{23b}\) (XXXIV) is a new fundamental type of the class
which is formed from geranyl farnesol (XXXIII) by a cyclisation
initiated at the isopropylidene groups.

![Chart - II](image-url)
The triterpenoids form a very large group of naturally occurring isoprenoids widely distributed throughout the plant kingdom. The triterpenoids ($\text{C}_{30}$) and the carotenoids ($\text{C}_{40}$) arise, not from continuous polymerization of $\text{C}_5$ units, but from specialized dimerisation of $\text{C}_{15}$ and $\text{C}_{20}$ units respectively, e.g. squalene (XXXV). The hexa-ene all-trans squalene (XXXV) is the immediate biological precursor of all triterpenoids. Squalene is transformed into lanosterol (an important compound of animal origin - XXXVI), cycloartenol (XXXVII), $\beta$-amyrin (XXXVIII), and various other triterpenoids and steroids via cyclisation of its terminal epoxide (XXXIX), or the hydrocarbon itself. Capstack et al. have demonstrated the direct cyclisation of squalene to $\beta$-amyrin with pea seed homogenates. For the relatively small number (about 40) of triterpenoids that are not oxygenated at $\text{C}_3$ it has been suggested that they arise from squalene by proton catalysed cyclisation (rather than formation, and subsequent cyclisation of terminal epoxide). Recently, Barton et al. and others have presented evidence for the above route (Chart - III) by biosynthetic experiments carried out for formation respectively of the pentacyclic hydrocarbons (e.g. fern-9-ene-XL) in P. vulgare, and tetrahymanol (XLI) from squalene. Squalene has been shown to be transformed into both fern-9-ene and $\beta$-sitosterol, but 2,3-oxidosqualene is converted only in the sterol.
CHART III

Fern-7-ene

Diploptene

(XL)

(XLI)

Hopene - 1
The Biogenetic Isoprene Rule postulated that the different types of tetra- and pentacyclic triterpenoids are formed according to the conformation that squalene (or, its epoxide) adopts, presumably at an enzyme surface prior to cyclisation. Each of the conformations leads stereospecifically to a particular cyclisation product. Only a few examples of the above may be cited in the present thesis because of the limited space.

Cyclisation of squalene-epoxide in the conformation (XLII) leads to the cation (XLIII) from which are derived some of the pentacyclic triterpenoids as shown in the Chart IV. The β-amyrin (Oleanane) group includes a very large number of naturally occurring pentacyclic members which cover an impressive variety of functional groups. There are at present known examples of oxygen functions attached to all nuclear positions in the oleanane skeleton except C7 and C11; of the angular methyl groups only that at C8 has not been found oxidised.

A great deal of chemical interest has in recent years25-27 centered also on the two groups known as limonoids (e.g. limonin - XLIV)25a,b,26 and quassinoids (e.g. quassin-XLV) which may be derived from euphol (XLVI) or tirucallol (XLVII). A number of nor-triterpenoid bitter principles related to quassin have also been isolated from plant sources25a,c,e,f recently.

The different routes through squalene cyclisation are impressive in their simplicity and generality, but certain other
proposals do exist\textsuperscript{10}. A large number of triterpenoids is also formed by secondary transformations of the preformed skeletons; thus, nyctanthic acid (XLVIII) contains the $\beta$-amyrin skeleton opened oxidatively\textsuperscript{24}, and limonin\textsuperscript{26} contains a much adapted skeleton. Another elegant modification is the presumed conversion of $\beta$-amyrin into the enol of friedelin (XLIX) via a backbone rearrangement\textsuperscript{27}. 

\begin{align*}
\text{(XLIV)} \\
\text{(XLV)} \\
\text{(XLVI) } 20\beta H \\
\text{(XLVII) } 20\alpha H \\
\text{(XLVIII)}
\end{align*}
The discovery of mevalonic acid\textsuperscript{28}(L) has proved of great significance in the study of the biogenesis and biosynthesis of natural substances which are derived, in part or completely, by the condensation of isoprenoid units. In fact, it has been shown that the Biogenetic Isoprene Hypothesis has its origin in acetic and mevalonic acids. Isopentane units may be derived from three molecules of acetic acid with the loss of a carboxylic acid group. Biosynthesis of squalene from mevalonate in mammalian liver has been carried out by Cornforth, Popjak and their collaborators. It is now clear that mevalonic acid (MVA) is the direct source of the isoprenoid units in terpene and sterol syntheses\textsuperscript{11d,29-33}, besides the evidence that MVA is the intermediate formed from acetate. It has been shown that MVA yields isoprene units by losing one carbon atom of its carboxylic acid group. Mevalonic acid has been obtained by condensation of acetyl-co-enzyme-A\textsuperscript{34}, and its status as an irreversible intermediate in isoprenoid biosynthesis has been revealed\textsuperscript{35}. 
Bloch, Lynen, and others have shown that isopentenylpyrophosphate (3-methyl-3-butenyl-1-pyrophosphate; A, Chart - V), obtained from labelled mevalonic acid as an intermediate is the building stone (active isoprene) for all naturally occurring isoprenoids. The compound A (Chart - V) is also converted by an enzyme to \( \gamma, \gamma \)-dimethylallyl pyrophosphate (3-methyl-2-butenyl-1-pyrophosphate; B - Chart - V). The units (A) and (B) then combine to produce geranyl pyrophosphate as first condensation product, which with another C_5 unit (A) gives farnesyl pyrophosphate. The latter is also then converted to squalene, which, subsequently on cyclisation, affords triterpenoids and steroids. The isomerisation of A to B is essential for biosynthesis of terpenoids to give their terminal isopropylidene group. In this way the biosynthetic steps from acetyl-co-enzyme-A to terpenoids, steroids and rubber are accomplished. It has been shown that the stereochemistry of elimination of hydrogen in the conversion of the unit A into the unit B (Chart - V) was the same in plant and animal tissue during biosynthetic experiments for squalene, \( \alpha \) - and \( \beta \) -amyrins and phytol (LI). Recent approaches have also elucidated the stereochemistry of the above condensation and dimerisation processes in an elegant manner. 
Mevalonic Acid → ATP → Mevalonic Acid-5-P → ATP → Mevalonic Acid-5-P-P

2TPNH → 2TPN⁺

β-Hydroxy-β-methyl-glutaryl-CoA

Acetoacetyl-CoA → Acetyl-CoA

Carbohydrates, Fats

2TPNH → 2TPN⁺

Steroids ← Triterpenoids ← Squalene

CO₂ + ATP

CH₂ = CH₂-CH₂-OP₂O₆H₃

Geranyl-P-P(₈C)

- H₄P₂O₇

Squalene ← TPNH + TPN⁺ (₈C₁₅) Farnesyl-P-P

Rubber

CH₃

CH₃-C=CH-CH₂[CH₂-C=CH-CH₂]ₙ C=CH-CH₂-O-P-O-P-OS⁻

CHART V
The radioactive forms of acetic or/and mevalonic acids have been converted into steroids, tri-, sester-, di-, sesqui-, and monoterpenoids. These acids have also been postulated to be precursors of isoprene units in the terpenoids of mixed biogenetic origin and non-terpenoid compounds. Some phenols, ubiquinones, plastoquinones, and vitamin K etc. have also been found to be of megalonoid origin. Other compounds of triterpenoid origin, e.g. vitamin A, trisporic acids, terpene alkaloids, cyclopropane compounds, and the fungal sesterterpenoids etc. have also been studied biosynthetically.

\[ \text{(LI)} \]

A detailed survey of biosynthesis and metabolism of monoterpenes has been published very recently by Banthorpe et al. Loomis and coworkers have shown that labelled glucose and CO\textsubscript{2} are more efficient precursors of monoterpenoids in peppermint cuttings than a mevalonate. It has also been shown by the same authors that when unlabelled sucrose is fed along with
labelled mevalonate, the incorporation of mevalonate into mono- and sesquiterpenoids is markedly increased\textsuperscript{50}.

Trichothecolone (LIII, \(R=H\)), the \(C_{15}\) moiety of the antifungal metabolite trichothecin (LIII, \(R=\text{isocrotyl}\)), is a sesquiterpenoid in origin as shown by biosynthesis\textsuperscript{43a} from mevalonic acid. Normal sesquiterpenoid skeleton can be achieved by a 1,3 or, double 1,2-methyl group migration as shown (LII \(\rightarrow\) LIII). Revised structure for trichothecolone is now LIII A. Biosynthesis of trichothecin and related compounds also has been attempted\textsuperscript{51}, and a number of approaches to the synthesis of hydroazulenic sesquiterpenes have been discussed recently\textsuperscript{9e}.
Biosynthetic type total synthesis of dl-tetrahymanol, \( \chi \)-amyrin, \( \beta \)-amyrin and germanicol have been accomplished by van Tamelen et al.\(^5\)\(^2\) The mode of hydride shifts\(^5\)\(^3\) accompanying 2,3-epoxysqualene cyclisation to lanosterol in yeast and to \( \beta \)-amyrin in peas has very recently been verified by Barton et al.\(^2\)\(^4\)\(^h\),\(^3\)\(^9\)

**STEROLS**

The sterols in general form a large group among natural compounds possessing a tetracyclic nucleus similar to that of lanosterol (XXXVI) and other tetracyclic triterpenoids, but only two methyl groups are attached to the ring system, at positions 10 and 13. The eight carbon side-chain found in lanosterol is also present in many steroids, especially from animal sources; but most plant steroids have one or two additional carbon atoms. All the steroids on selenium dehydrogenation yield among other products Diel's hydrocarbon (LIV). A steroid may, therefore, be defined as any compound which yields Diel's hydrocarbon on selenium dehydrogenation. Steroids occur in a wide variety of organisms in the animal as well as plant kingdom. The name 'sterol'\(^1\)\(^a\) applies specially to steroid alcohols; but, since practically all plant steroids are alcohols with a hydroxyl group at C-3, they are frequently all called 'sterols'. 
Steroids occur throughout the plant kingdom as free sterols and their esters. In plant they are said to have no known function although they have profound importance in animal metabolism as hormones, coenzymes, bile acids and pro-vitamin D etc. The role of sterol-esters in plants also does not appear to be very clear. Certain animal steroids have been shown to influence plant growth strongly.

Pregnane-type steroids (e.g. $\Delta^5$-pregnenol-3$\beta$-one-20 — LV) and cholesterol (LVI) have recently been found in plants as well. Steroids of zymosterol (LVII) type are known in yeast, fungi and algae, but their presence have hardly been established in higher plants. The stigmastane-type (e.g. stigmasterol-LVIII; $\alpha$-spinasterol-LIX; $\beta$-sitosterol-LX; and campesterol-LXI etc.) is most characteristic in higher plants. Other sterols differing in the position and number of double bonds are also reported in different plant material. A list of 55 natural steroids of stigmastane type, supposed to be known so far, has been given elsewhere.
It appears difficult at present to draw any definite conclusions with regard to the taxonomic distribution of the various sterols. It may, however, be concluded\(^\text{53}\) that ergosterol (LXII) is the principal sterol of fungi, and that cholesterol and fucosterol (LXIII) are the typical sterols of majority of Rhodophyceae and Phaeophyceae respectively. Most higher plants contain predominantly 24\(\alpha\)-sterol (usually \(\beta\)-sitosterol) with a few exceptions. Some 4\(\alpha\)-methyl sterols\(^\text{55}\) (e.g. Lophenol-LXIV; and cycloeucalenol-LXV) also have been isolated and identified from a number of plant sources. Acansterol\(^\text{56a}\)(LXVI) is a cyclopropane containing sterol, and a number of similar sterols have been isolated and identified from red alga porphyridium cruentum\(^\text{56b}\) and C. emergonii\(^\text{56c}\). Barton et al.\(^\text{57}\) have recently isolated and identified from yeast a large number of sterols including fecosterol (LXVII), episterol (LXVIII), derivatives of lanosterol, and zymosterol besides those which were previously unreported in yeast so far (e.g. parkeol-LXIX etc.).
The biogenetic relationship between squalene (LXX), lanosterol and cholesterol has been widely studied\(^{29-33,39}\). More recent work has clarified the nature of the electrophilic attack that initiates cyclisation of squalene 2,3-oxide (LXXI). A rat liver preparation has been converted into lanosterol almost quantitatively\(^{58}\). It is considered that cyclisation is triggered by the protonation and opening of the epoxide ring (Chart - VI) such that the 4α-methyl group is derived from the C2 of MVA\(^{59a}\). Several pathways have been suggested for the conversion of lanosterol into cholesterol. It is suggested that C14 methyl group is probably removed before the gem-dimethyls by means of this sequence: hydroxymethyl-aldehyde-acid, followed by decarboxylation\(^{6c,59b}\). Cholesterol is mentioned to be the precursor for other steroids, sex hormones etc.
Incorporation of acetate and mevalonate into \( \beta \)-sitosterol, acidic pentacyclic triterpenes, and squalene has been observed by Nicholas.\(^{60}\) It has also been shown that mevalonate was incorporated into \( \beta \)-amyrin, \( \beta \)-sitosterol, and other phytosterols.\(^{61}\) The incorporation of acetate, mevalonic acid and squalene into ergosterol by yeast is well established. It has also been shown that generally phytosterols arise from mevalonate and squalene by a biosynthetic pathway very close to that of cholesterol.\(^{24b,62,63}\)
Although lanosterol has been isolated from microorganisms and a few plants but evidence has been shown that it is not on the direct route to phytosterols according to Goad. Cycloartenol (XXXVII) which occurs widely and was rapidly labelled with tracer from acetate or MVA in plants and tissue cultures under conditions where lanosterol could not be detected, appears to take its place. Squalene 2,3-epoxide was converted into cycloartenol but not into lanosterol, and the cyclopropane compound could be further converted into other phytosterols. The presence of lanosterol in latex from certain species has been interpreted as the result of enzymic modification of cycloartenol.

Cycloartenol can be hypothetically derived from the c-b-c-b-u conformation of squalene via the intermediate (LXXII), and the evidence for the hydrogen migration has been provided by feeding experiments.
However, some species appear capable of transforming lanosterol into phytosterols $^{66a}$. The sequence from cycloartenol to other phytosterols is not very clear, although various experiments to explore precursor relationship have been carried out$^{62,69-71}$. van Tamelen et al.$^{72}$ have recently carried out biosynthetic type total syntheses of a number of sterols including isoeuphenol (LXXIII) and parkeol etc. Biosynthetic experiments for sterols have also been recently carried out by a number of other workers$^{73,74}$. Barton et al.$^{30a}$ have presented a data showing that the squalene-epoxide route is applicable also to yeast sterols. Preliminary studies on the biosynthesis of ergosterol in yeast also have been carried out and it has been demonstrated that ergostatetraenol (LXXIV) is a precursor for ergosterol in yeast$^{75}$.

Castle et al.$^{76}$ have suggested the alkylation mechanism of phytosterol side-chain as outlined in Chart - VII. It is envisaged that a $\Delta^{24}$-steroid intermediate (LXXV) is methylated by methionine to give a carbonium ion (LXXVI) which may be stabilised either by the addition of a hydride ion (route a) to give a 24-methyl steroid (LXXVII), or by loss of a proton from the introduced methyl group (route b) to leave a 24-methylene steroid (LXXVIII). In the latter case a second transmethylation will produce a further carbonium ion (LXXIX) which can then give
either a 24-ethyl (LXXX) or a 24-ethyldiene (LXXXI) steroid (routes c and d). If the loss of a proton (route d) is a stereospecific process then such a scheme could explain the formation of fucosterol (LXIII) or 29-iso-fucosterol (LXXXII) in various plants. In short, all experimental evidence is considered to be in accord with the intermediate production of 24-methylene or ethyldiene sterols followed by reduction to give the 24-methyl or 24-ethyl sterols respectively. According to Goad and Goodwin$^{69}$ and also Lederer$^{77}$ the C-alkylation of the side chain of ergosterol etc. can probably occur at the C-27, C-30 or any intermediate structural level by a route of methyl transfer from S-adenosyl-methionine$^{63b}$, and migration of hydrogen from C-24 (route e). Further alkylation and reduction of the methylene sterols formed can subsequently occur (routes f and g).
Formation of various other types of sterols (e.g., 24-methylene-lophenol - LXXXIII and macdougallin - LXXXIV) has also been studied by different workers. 24,25-Dihydro-24-methylene lanosterol has been synthesized and its role (as a precursor) in the biosynthesis of steroids and triterpenoids (C-alkylation of side chain) has been investigated with respect to eburicoic acid (LXXXV) and ergosterol by Barton et al. Their results give support to the hypothesis of alkylation at the lanosterol stage and indicate that yeast can assimilate a series of 24-methylene sterols. According to their findings the above compound has been excluded as a precursor of ergosterol in Saccharomyces cerevisiae.
ACETOGENINS

The conversion of acetate to the acetogenins* is a process that possesses a close similarity to the construction of fatty acids. The acetate hypothesis (first suggested by Collie) states that a linear polyketomethylene chain, \( \text{CH}_3\text{CO-}(\text{CH}_2\text{CO})_n\text{-CH}_2\text{-COOH} \), formed from head-to-tail self-condensation of acetate units could cyclise into a remarkable array of complex structures. New carbon-carbon bonds are formed by the addition of a malonyl coenzyme-A to an activated carbonyl group. Malonyl coenzyme-A is itself formed from acetate by carboxylation of acetyl coenzyme-A.

\[
\begin{align*}
\text{CH}_3\text{COSCoA} + \text{CO}_2 & \rightarrow \text{CH}_2\text{COSCoA} \\
& \quad \quad \text{COOH}
\end{align*}
\]

\[
\begin{align*}
\text{CH}_3\text{COSCoA} + \text{CH}_2\text{-COSCoA} & \rightarrow \text{CH}_3\text{C-CH}_2\text{-COSCoA etc.} \\
& \quad \quad \text{COOH}
\end{align*}
\]

An unreduced polyketo-chain may be considered as an intermediate that can undergo internal, aldol-type reactions to yield cyclic products (e.g. Orsellinic acid – LXXXVI). The oxygen functions in these compounds (acetogenins) frequently marked the carboxyl

carbons of acetate units and serve as an important key to the structural deduction of acetate biogenesis. An additional oxidation (or reduction) may also take place to yield products in different over-all oxidation states from those of the formal polyketo-chain precursors (e.g. Cyclopaldic acid - LXXXVII). Other modifications and various rearrangements may afford different types of compounds belonging to the family of acetogenins.

An enolate ion may also attack a keto-group at another portion of the chain to yield cyclised products of either 0 or C acylation. These reactions occur with facility in forming six-membered rings, the C-alkylation (Aldol or Claisen condensation) in this case leading to a phenolic aromatic ring. Two major routes (paths A and B) lead to two families of phenols, the acylphloroglucinol (LXXXVIII) and Orsellinic acid derivatives (LXXXIX) respectively, distinguished by their aromatic substituent patterns. Reduction of an uninvolved ketone before cyclisation can lead analogously to the generalised types (XC) and (XCI).

The terminal acid of the polyacetyl chain \( \text{CH}_2\text{COOH} \) may not always be acetic acid. It is considered that another acid \( R\text{COOH} \) might initiate the extension of a chain with malonyl units. For an instance shikimic-derived acid (CXII) may be chain-extended by malonyl units, similar to acetic acid, to provide polyacetyl chain with aromatic terminals. It has been
\[
\begin{align*}
\text{CH}_3\text{COSCoA} & \xrightleftharpoons{-3\text{CO}_2} 3\text{CH}_2-\text{COSCoA} \\
\text{etc.} & \quad \text{(LXXXVI)}
\end{align*}
\]
suggested that large number of benzenoid compounds in nature almost invariably derive their aromatic rings by one of the two routes\textsuperscript{93}: (i) via shikimic acid (XCI\textsubscript{II}), or (ii) through poly-acetyl chain cyclisation. The biogenesis of shikimic acid as examined experimentally may be shown (Chart - VIII).

<table>
<thead>
<tr>
<th>Prephenic Acid</th>
<th>(XCIV)</th>
<th>p-Hydroxyphenyllactic Acid (XCVIII)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Phenylpyruvic Acid</td>
<td>(XCV)</td>
<td>Tyrosin (XCIX)</td>
</tr>
<tr>
<td>Phenylalanine</td>
<td>(XCVl)</td>
<td>p-Coumaric Acid (C)</td>
</tr>
<tr>
<td>Cinnamic Acid</td>
<td>(XCVII)</td>
<td>Caffeic Acid (CI)</td>
</tr>
</tbody>
</table>
CHART VIII
In order to verify the acetate hypothesis regarding the formation of acetogenins 6-methyl salicylic (CII) and orsellinic (LXXXVI) acids, as first examples of acetogenins, have been examined by biosynthetic experiments from labelled acetic acid and the labelling pattern in these compounds has been found to be in conformity with the prediction of the acetate hypothesis. Lynen has shown that an enzyme extract can catalyse the reaction of one acetyl coenzyme-A and 3-malonyl coenzyme-A molecules to produce 6-methyl salicylic acid. The orsellinic origin of cyclopaldic acid (LXXXVII) has also been confirmed by similar experiments. Besides such several experiments regarding formation of different aromatic compounds, citrinin (CIII) biosynthesis has also been studied which confirms the acetate origin of the nucleus and the latter has shown that labelled methionine (CIV) was more efficient precursor than formate of the two extra methyl groups and the carboxyl. Labelled acetic acid was also incorporated into the complex acetogenins fuscin (CV), griseofulvin (CVI) and various other compounds of the type.

Birch et al. have made investigations on the chemistry of poly-β-ketones as models for the polyacetyl chain, and the Collie's experiments with diacetyl-acetone (CVII) have also been confirmed. Using the phenyl-terminated polyacetyl derivative (CVIII) they were able to synthesize the natural dihydropinosylvin (CIX) confirming the capability of polyacetyl chains to undergo
the postulated and often complex condensations of the acetate hypothesis.

Flavonoids and coumarins etc. (which make a part of the present work) have been classified as 'Acetogenins'; hence, some aspects of flavonoid chemistry may also be discussed in the following pages.

**FLAVONOIDS**

The flavonoids $^7b,c,10a$ belong to a widely distributed group of plant pigments and are found practically in all parts of plants. They, in their occurrence, represent a very large number of types with different properties, and are characterised by a Ph-C$_3$-Ph skeleton (CX). These compounds occur mostly as glycosides (e.g. triglycoside - Robinin; diglycoside - butrin; and monoglycoside - quercitrin), and their various forms are distinguished by the functionality and state of oxidation of the central C-3 chain (Chart IX for a few examples).
Flavones - (CXI)
Flavanones (CXII)
Isoflavones (CXIII)

CHART IX
Besides qualitative tests, colour reactions and chromatographic techniques, spectrophotometric measurements are now commonly employed in the characterisation of flavonoids. The value of spectral data in the identification and structural elucidation of these plant pigments has been increased considerably by the use of reagents such as aluminium chloride, sodium ethylate, fused sodium acetate, and boric acid-sodium acetate which produce shifts in the maxima in the U.V. spectra in accordance with the location of the various functional groups in the flavonoid molecule (CXIV). Sometimes, a flavonoid may be submitted to degradation processes (e.g. alkaline hydrolysis) and the fragments are identified. The structure is then deduced, and finally confirmed, if possible, by synthesis.

It has generally been presumed that the entire flavonoid group of natural products arises by an acylphloroglucinol cyclisation of a polyacetyl-chain with one phenyl-propionic acid as terminal (CXV). The flavonoid A rings generally display oxygen at both positions 5 and 7 (or, one of these if only one oxygen), and the B rings the typical shikimic-derived patterns of ring hydroxylation. Some compounds also carry C-alkylations (e.g. methyl, methylene, isopentenyl, or C-glycosides — e.g. Orientin — CXVI) and the latter are generally found on the A rings (at position 6 or 8). A large number of bis-flavones[89] (Biflavonoids) are also now known illustrating oxidative coupling of two flavonoid
residues (e.g. Gingketin - CXVII).

The production of many derivatives of orcinol and phloroglucinol and some larger phenolic molecules was postulated through head-to-tail linkage of acetic acid units, and the generality of this hypothesis has been confirmed by several
biochemical experiments. Phloroglucinol ring has been postulated according to the following scheme (Chart I) where R can correspond to any acid normally found in nature. Various other schemes and ideas have also been proposed for flavonoid biosynthesis, e.g., starting from cinnamic acid (or related compound) derived by shikimic acid route with the addition of three acetic acid units (Chart - VIII). Certain experiments have been performed using labelled precursors related to the shikimic acid-phenylalanine group, and acetic acid. Labelled shikimic acid (XClII), p-coumaric acid (C) phenylalanine (XCVI) or cinnamic acid were all incorporated into the C⁶-C₃ portion (B) of quercetin⁹¹(CXVIII). The C₃ portion was incorporated intact, and caffeic acid (CI) occurring with quercetin was found to be active. Good precursors of ring B were shikimic acid, phenylalanine, cinnamic and p-hydroxy-cinnamic acids, but p-hydroxybenzoic, 3,4-dihydroxy-benzoic and caffeic acids were poor. Ring A of quercetin and also of cyanidin⁹² (CXIX) has conclusively been shown to arise from acetic acid. Labelled acetate yielded also phloroglucinol. The use of labelled glucose, a source of shikimic acid and also of active acetic acid (through pyruvic acid), gave almost uniformly labelled quercetin.
However, it has been found that the fungal terphenyl, volucrisporin (CXX), is labelled by exogenous phenylalanine, m-tyrosin, or shikimic acid but not by acetate. Flavonoidal compounds may be formed from phenylpyruvic acid (XCV). It has been observed that phenylpyruvic acids themselves have not
been found in plants, but recent evidence supports a route in which this key intermediate is most commonly converted first to phenylalanine, which in turn serves as the general precursor of phenylpropanoids (via cinnamic acid — Chart — VIII).

Fungenin, a glucoside of 3,4-dihydroxyacetophenone, has been shown to have a shikimic origin; the best precursor were phenylalanine and caffeic acid, whereas labelled cinnamic, p-coumaric, phenyllactic and shikimic acids were also good, but acetic acid was very poorly incorporated.

Coumarin (CXXI) has been variously examined with the observations that labelled shikimic acid, phenylalanine, cinnamic, and o-coumaric acids were also incorporated; however, labelled acetate incorporation was very poor. The more complex coumarin, scopoletin (CXXII), has been investigated and it has been shown that phenylalanine is an excellent precursor. The fungal coumarin, novobiocin (CXXIII), was also shown to arise from labelled tyrosine, which is incorporated as a whole into the coumarin portion of novobiocin.