RAFFLES INSTITUTION
2016 YEAR 6 PRELIMINARY EXAMINATION

Higher 3

PHARMACEUTICAL CHEMISTRY 9812/01

Paper 1

23 September
2 hours 30 minutes

Additional Materials: Answer Paper
Data Booklet

READ THESE INSTRUCTIONS FIRST

Write your index number, civics tutorial group and name on the Cover Page.
Write in dark blue or black pen on both sides of the writing paper.
You may use a soft pencil for any diagrams or graphs.
Do not use staples, paper clips, highlighters, glue or correction fluid.

Answer any **five** questions.
Begin each question on a fresh sheet of paper.
At the end of the examination, fasten all your work securely together, with the cover page on top.

The number of marks is given in brackets [ ] at the end of each question or part question.
You may use a calculator.
You are reminded of the need for clear presentation in your answers.

This document consists of **21** printed pages.

1 Analgesic drugs are a diverse group of compounds that are capable of relieving pain. They can be classified into two categories, the narcotic and non-narcotic analgesics.

(a) Narcotic and non-narcotic analgesics act differently to prevent pain. Briefly describe the mode of action of the narcotic analgesics. [1]

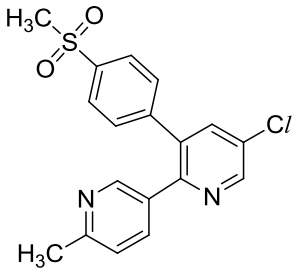
(b) Non-steroidal anti-inflammatory drugs (NSAIDs) belong to the class of non-narcotic analgesics. NSAIDs are inhibitors of cyclooxygenase (COX-1 and COX-2).

Traditional NSAIDs, such as aspirin, are considered non-selective because they inhibit both COX-1 and COX-2.

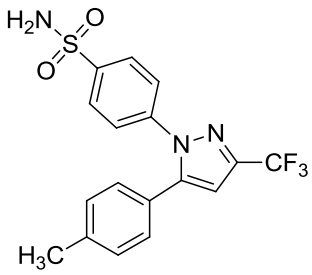
The inhibition of COX-1 leads to NSAID toxicity and associated side effects.

(i) List two side effects of aspirin.

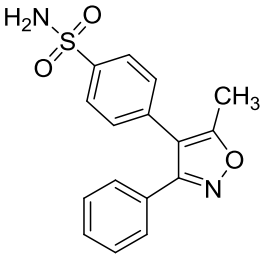
The inhibition of COX-2 accounts for the anti-inflammatory effect of NSAIDs. The following NSAIDs (coxibs) selectively inhibit COX-2.



etoricoxib



celecoxib



valdecoxib

(ii) The COX-2 active site is significantly larger than that of COX-1. Explain how the larger active site of COX-2 helps in the design of COX-2 selective inhibitors.

(iii) Suggest two structural features which allow the above coxibs to be selective COX-2 inhibitors.

The heterocyclic rings found in etoricoxib, celecoxib and valdecoxib are as shown:

pyridine

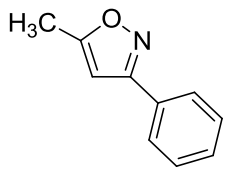
pyrazole

isoxazole

(iv) With the aid of resonance structures, explain why pyridine is more resistant to electrophilic substitution than benzene.

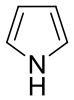
(v) Draw the structures of the cations derived from the protonation of pyrazole and isoxazole. Hence, explain whether pyrazole or isoxazole is more basic.

(vi) The substituted isoxazole shown below can be synthesised by reacting NH_2OH with a diketone.



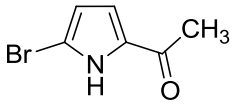
Identify the diketone for the above synthesis and suggest a possible side product.

Pyrrole is structurally similar to pyrazole and is used as a starting compound in organic synthesis.



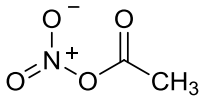
pyrrole

(vii) Propose a 2-step synthesis of 2-acetyl-5-bromopyrrole from pyrrole. State clearly the reagents and conditions required for each step.



2-acetyl-5-bromopyrrole

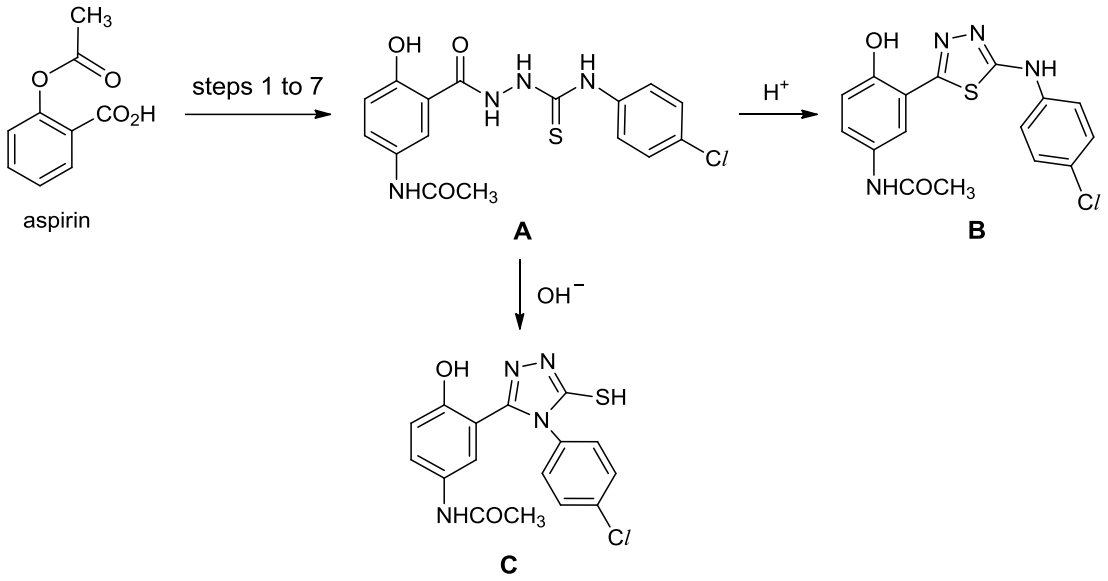
(viii) Outline the mechanism for the nitration reaction between pyrrole and acetyl nitrate.



acetyl nitrate

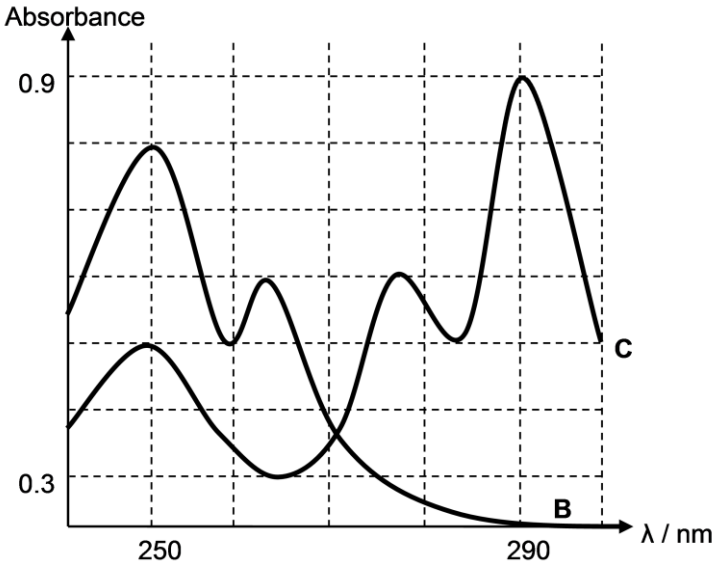
[14]

(c) Aspirin, a non-narcotic analgesic, can be used to synthesise anti-fungal drugs containing heterocyclic rings such as thiadiazole in **B** and triazole in **C**. The following scheme shows part of the synthesis of compounds **B** and **C** from aspirin.



(i) Explain why compound **A** undergoes two different reactions to form compounds **B** and **C** under different pH conditions.

The UV/Vis spectra of compounds **B** and **C**, each at 3.0 x 10^{-5} mol dm^{-3}, in a cell of path length 1.0 cm are shown below:



(ii) Explain why λ_{max} of compound **C** occurs at a longer wavelength than that of compound **B**.

In a clinical trial, a patient suffering from fungal infection was treated with a mixture of **B** and **C**. After 5 hours, a urine sample was collected and analysed with UV/Vis spectroscopy at 250 nm and 290 nm, in a cell of path length 1.0 cm:

Wavelength / nm	Absorbance
250	0.92
290	0.50

(iii) Determine the concentration of compound **C** in the urine sample.

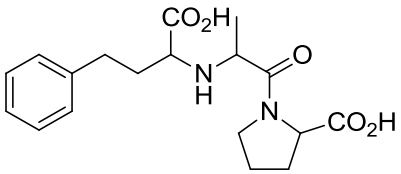
(iv) If the molar extinction coefficient of compound **C** at 250 nm is 14900 mol^{-1} dm^3 cm^{-1}, calculate the absorbance of compound **B** in the urine sample and hence determine its concentration.

[5]

[Total: 20]

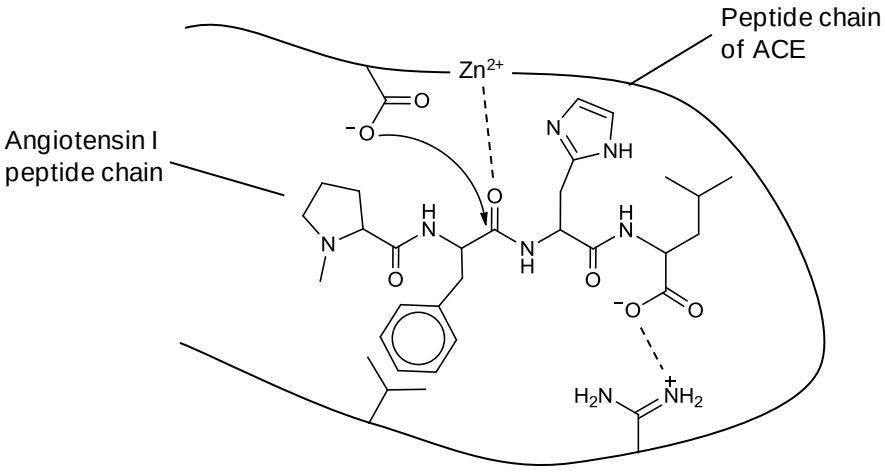
2 Angiotensin-converting enzyme (ACE) inhibitors are used for the treatment of hypertension (high blood pressure) and congestive heart failure. ACE inhibitors block the body's production of angiotensin II, a hormone that constricts blood vessels.

Enalapril is a prodrug that undergoes hydrolysis to form enalaprilat, a competitive inhibitor of ACE.



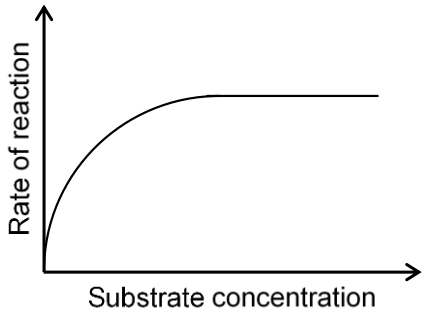
enalaprilat

(a) (i) ACE catalyses the conversion of angiotensin I to angiotensin II by hydrolysing the peptide bond between Phe and His. Angiotensin I binds to ACE in the model shown below.



By considering the structure of enalaprilat, draw a diagram to show and explain how enalaprilat acts as a competitive inhibitor of ACE.

The graph below shows how the initial rate of an enzyme-catalysed reaction varies with substrate concentration.



(ii) Explain the variation shown, using the idea of active sites.

(iii) Redraw the graph and, on the same axes, show how the initial rate would vary with substrate concentration in the presence of a reversible

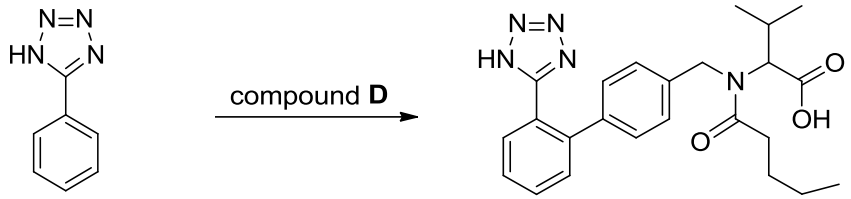
I: competitive inhibitor
II: non-competitive inhibitor

Label your graphs clearly.

[7]

(b) Due to the side effects of many ACE inhibitors, they have been widely replaced with angiotensin II receptor antagonists. An example is valsartan.

The synthesis of valsartan involves a reaction between compound **D** and 5-phenyl-1*H*-tetrazole.



5-phenyl-1*H*-tetrazole valsartan

Compound **D** has M_r = 384. In the mass spectrum of compound **D**, there are two peaks at *m/e* = 384 and 385 with a ratio of 5:1. There are two pairs of peaks of roughly equal intensity: one pair at *m/e* = 269 and 271, and another at *m/e* = 227 and 229.

The NMR spectrum data of compound **D** is given in the table below.

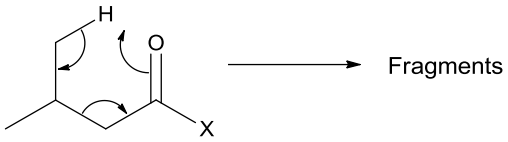
δ (in ppm)	Integration	Splitting
0.90	9	m
1.31	2	m
1.53	2	m
2.05	2	t
3.06	1	m
3.68	3	s
4.41	1	d
4.46	2	s
7.12	2	d
7.85	2	d

(s = singlet; d = doublet; t = triplet; m = multiplet)

(i) Using the spectra data, work out the molecular formula of compound **D**.

(ii) Draw the structure of **D** and account for the NMR peaks at δ (in ppm) 3.68, 7.12 and 7.85.

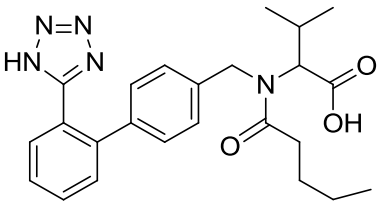
(iii) Fragmentation of molecular ions in the mass spectrometer is sometimes accompanied by the McLafferty rearrangement as shown below:



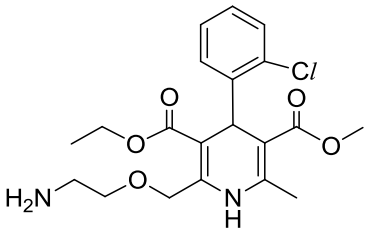
Identify the fragments responsible for the peaks at *m/e* = 269 and 227.

[9]

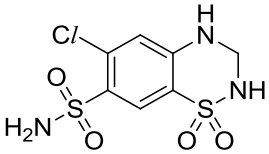
(c) Drugs formulated to treat hypertension usually contain a mixture of valsartan (VAL), amlodipine (AML) and hydrochlorothiazide (HCT).



valsartan
(VAL)



amlodipine
(AML)



hydrochlorothiazide
(HCT)

The composition of such drugs can be easily analysed by TLC. The following steps describe such a method.

1. A known mass of the drug was dissolved in methanol and applied on a silica gel plate.
2. The separation was carried out using a mixture of ethyl ethanoate, toluene and methanol in the proportions of 2:1:1 by volume. The plate was developed over a distance of 8 cm.
3. The positions of the spots of the three separated compounds were detected.

The R_f values are: AML, 0.42; HCT, 0.68; VAL, 0.19.

(i) Draw to scale, a diagram of the completed chromatogram, showing the positions of the spots of each compound. Include the positions of the start line and solvent front on your diagram.

(ii) How can the areas of the chromatogram containing each compound be identified in step 3?

(iii) Which of the compounds, AML, HCT or VAL, is the most polar? Briefly justify your answer from their R_f values.

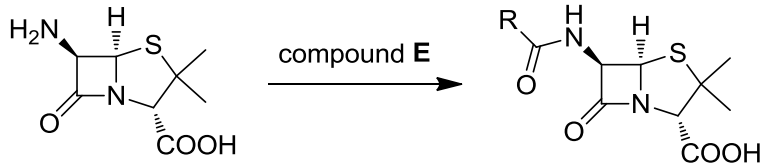
(iv) Deduce how the R_f value of AML will change if ethanoic acid was added to the mobile phase.

[4]
[Total: 20]

3 Penicillin is a widely used antibiotic which works by inhibiting peptidoglycan synthesis in the cell walls of Gram-positive bacteria.

(a) Apart from the action of penicillin, outline two other ways in which antibiotics act. [2]

(b) 6-aminopenicillanic acid is used as the main starting block for the preparation of numerous semisynthetic penicillin (e.g. penicillin G, penicillin V).



6-aminopenicillanic acid

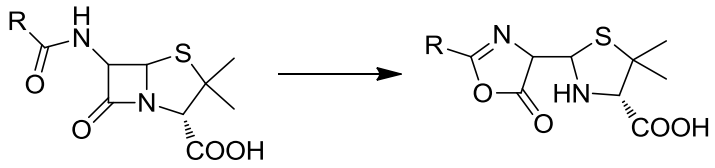
penicillin G (R =)

penicillin V (R =)

(i) Suggest the structure of compound **E** for the synthesis of penicillin G.

(ii) The β-lactam ring is an essential structure which confers the antibiotic properties of penicillin. However, penicillin is generally sensitive to acids, and is often not prescribed orally.

Outline the mechanism of the following reaction of penicillin under acidic conditions.

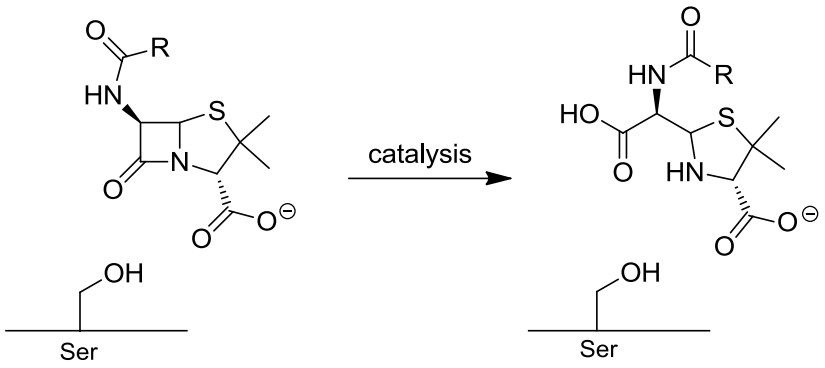


(iii) Penicillin V is found to be a more stable derivative than penicillin G, and hence it can be administered orally. Suggest a reason why this is so.

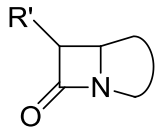
[4]

(c) In the 1960s, due to the prevalent use of penicillin, 80% of all *S. aureus* infections in hospitals were found to be caused by penicillin-resistant strains. These penicillin-resistant bacteria produce β-lactamase enzymes which catalyse the ring opening reaction of the β-lactam in penicillin.

The active site of β-lactamase contains a serine residue which forms a stable adduct with penicillin before the β-lactam ring is subsequently hydrolysed.



(i) Outline the mechanism of the hydrolysis of penicillin catalysed by β-lactamase. In your answer, you may represent penicillin as



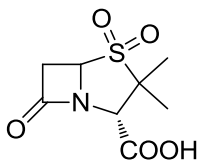
(ii) Explain how the failure to complete the prescribed course of antibiotics results in an increasing trend of bacterial resistance.

(iii) Suggest one modification to increase the efficacy of penicillin by reducing its susceptibility towards hydrolysis by β-lactamase.

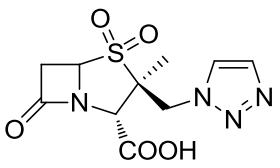
[6]

(d) Due to the prevalence of penicillin-resistant bacteria, β-lactamase inhibitors have been chemically synthesised and administered together with penicillin.

These β-lactamase inhibitors work by reacting with β-lactamase. Two examples of β-lactamase inhibitors are shown below.



sulbactam

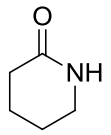


tazobactam

(i) Apart from the β-lactam ring, suggest one similarity and one difference present in the two β-lactamase inhibitors compared to penicillin.

(ii) Suggest how these two β-lactamase inhibitors can be distinguished by their infra-red (IR) spectra.

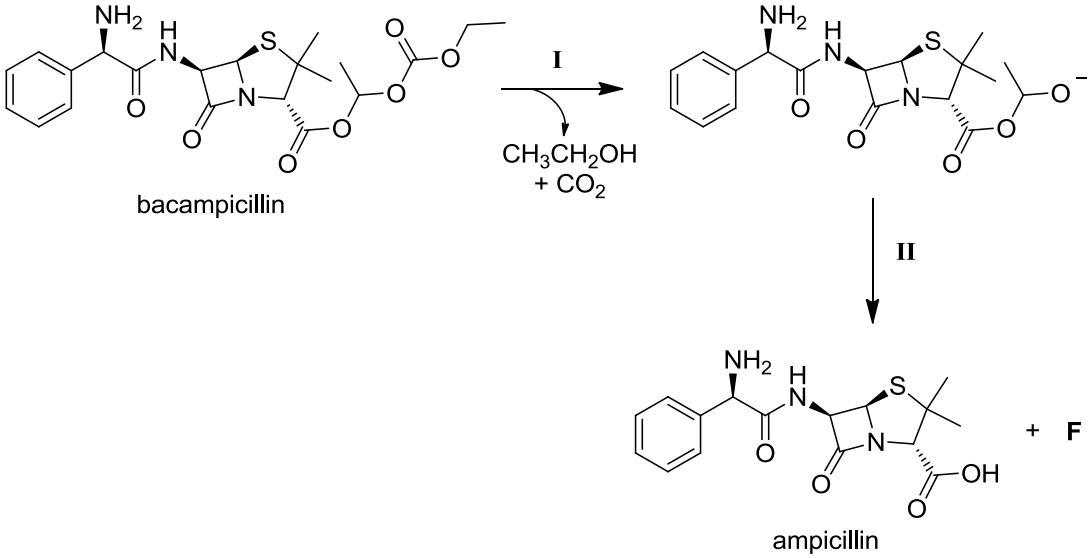
(iii) Predict if the wavenumber of the C=O stretch present in the β-lactam ring is higher or lower than that of the C=O stretch present in δ-lactam ring. Explain your answer.



δ-lactam ring

[5]

(e) Ampicillin was one of the most widely used broad spectrum penicillin in the 1960s. Bacampicillin, the prodrug of ampicillin, is often administered orally.



(i) Explain how the use of bacampicillin as a prodrug makes treatment with ampicillin more effective.

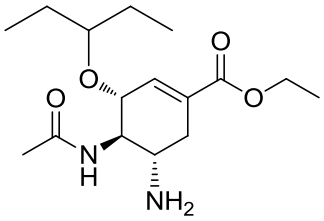
Reaction **I** is catalysed by the enzyme, esterase, present in the intestinal walls, while reaction **II** is spontaneous. Compound **F** is produced as an organic by-product.

(ii) Suggest the identity of **F**.

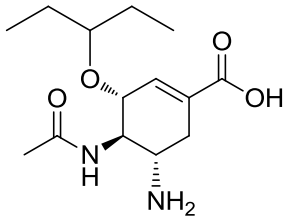
[3]

[Total: 20]

4 Tamiflu® is an antiviral drug used in the treatment of both influenza virus A and influenza virus B. It contains oseltamivir in the form of oseltamivir phosphate. Oseltamivir is an ethyl ester prodrug that is hydrolysed *in vivo* by hepatic esterases to its active form, oseltamivir carboxylate. Oseltamivir carboxylate has high bioavailability and penetrates sites of infection at concentrations that are sufficient to inhibit viral replication.

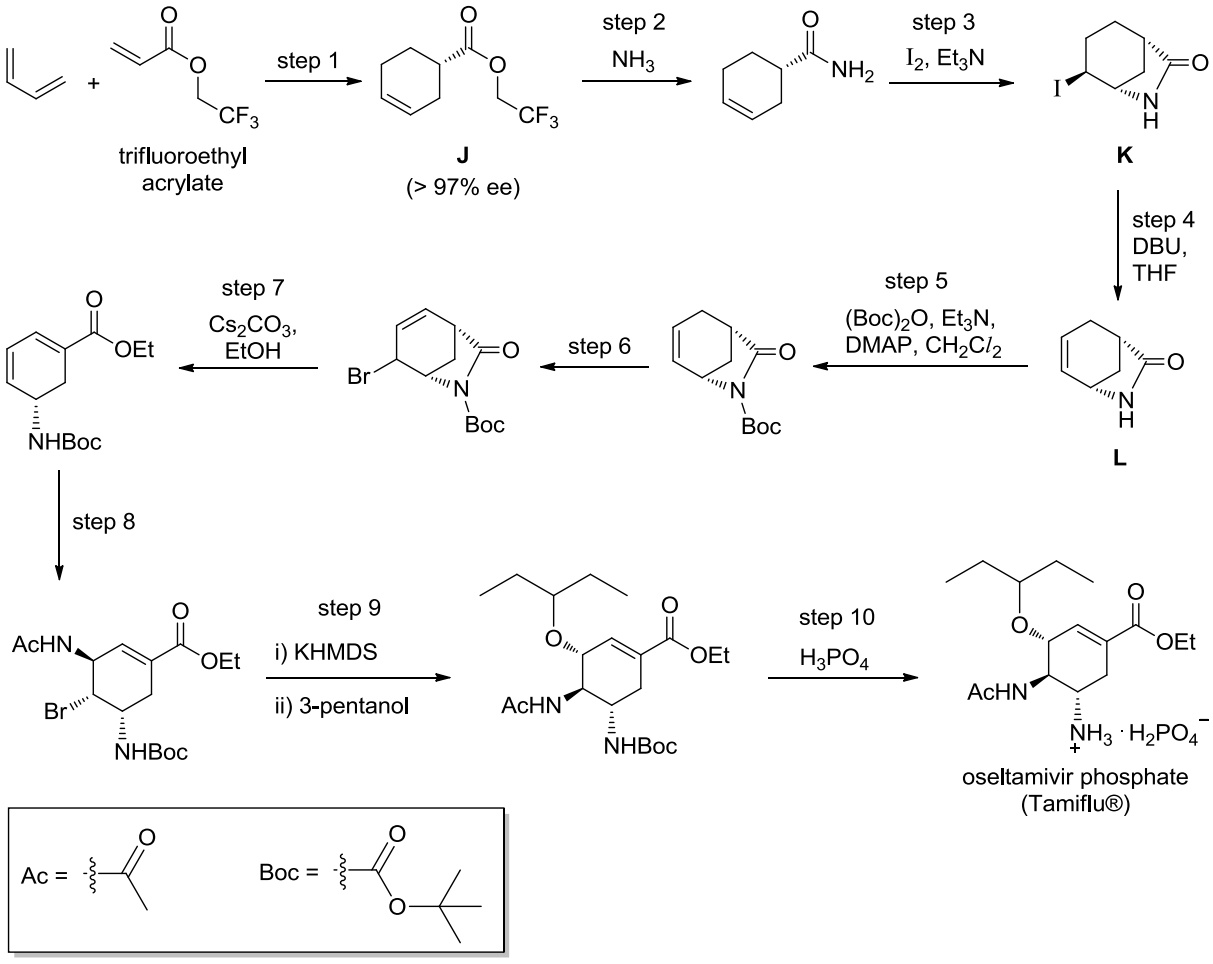


oseltamivir



oseltamivir carboxylate

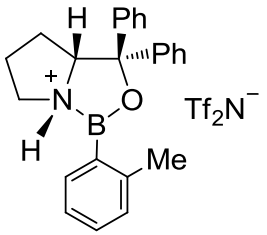
In 2006, E. J. Corey and his group published a novel total synthesis of oseltamivir phosphate starting from butadiene and trifluoroethyl acrylate as shown below:



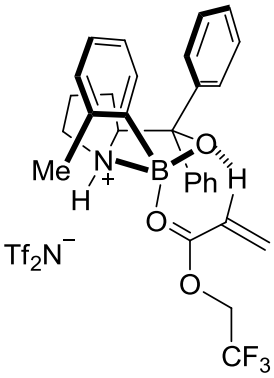
(a) Step 1 is an asymmetric Diels-Alder reaction to form compound **J**.

(i) Given that the reaction proceeds via a one-step concerted mechanism, suggest the mechanism for step 1. You may ignore the stereochemistry of the product in your answer.

(ii) The enantioselectivity of this reaction is made possible with the use of a (*S*)-proline-derived catalyst. The catalyst forms an adduct with the trifluoroethyl acrylate, which then undergoes an asymmetric Diels-Alder reaction with butadiene.



S-proline-derived catalyst



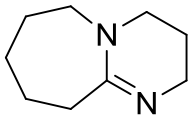
Adduct formed between catalyst and trifluoroethyl acrylate

Based on the structure of the above adduct, suggest two reasons how it allows the Diels-Alder reaction to be enantioselective.

[3]

(b) Using stereochemical projections, outline the mechanism for step 3 to account for the stereochemistry of **K**. [2]

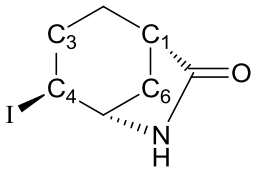
(c) Step 4 involves an E2 reaction using 1,8-Diazabicyclo[5.4.0]undec-7-ene (DBU) as the base.



DBU

(i) Suggest two reasons why aqueous NaOH should not be used in step 4.

(ii) By drawing Newman projection through C4-C3 and C6-C1 bonds of the cyclohexane ring in **K**, outline the mechanism for the conversion of **K** to **L**. In your answer, you may abbreviate DBU as **B**.



K

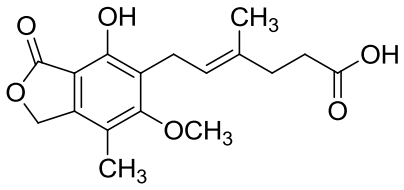
[5]

(d) Potassium bis(trimethylsilyl)amide (KHMDS) used in step 9 is a strong base.

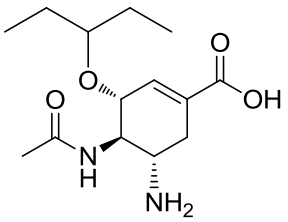
Given that step 9 involves two successive nucleophilic substitution (S_N2) reactions, outline the mechanism for step 9. In your answer, use appropriate stereochemical projections to account for the stereochemistry of the product. You may abbreviate KHMDS as **B**. [4]

Recent studies have highlighted that influenza patients who took Tamiflu®, especially those aged between 10 to 17 years old, seemed to be at higher risk of showing abnormal behaviour, delirium and hallucination, compared to those who did not take the drug.

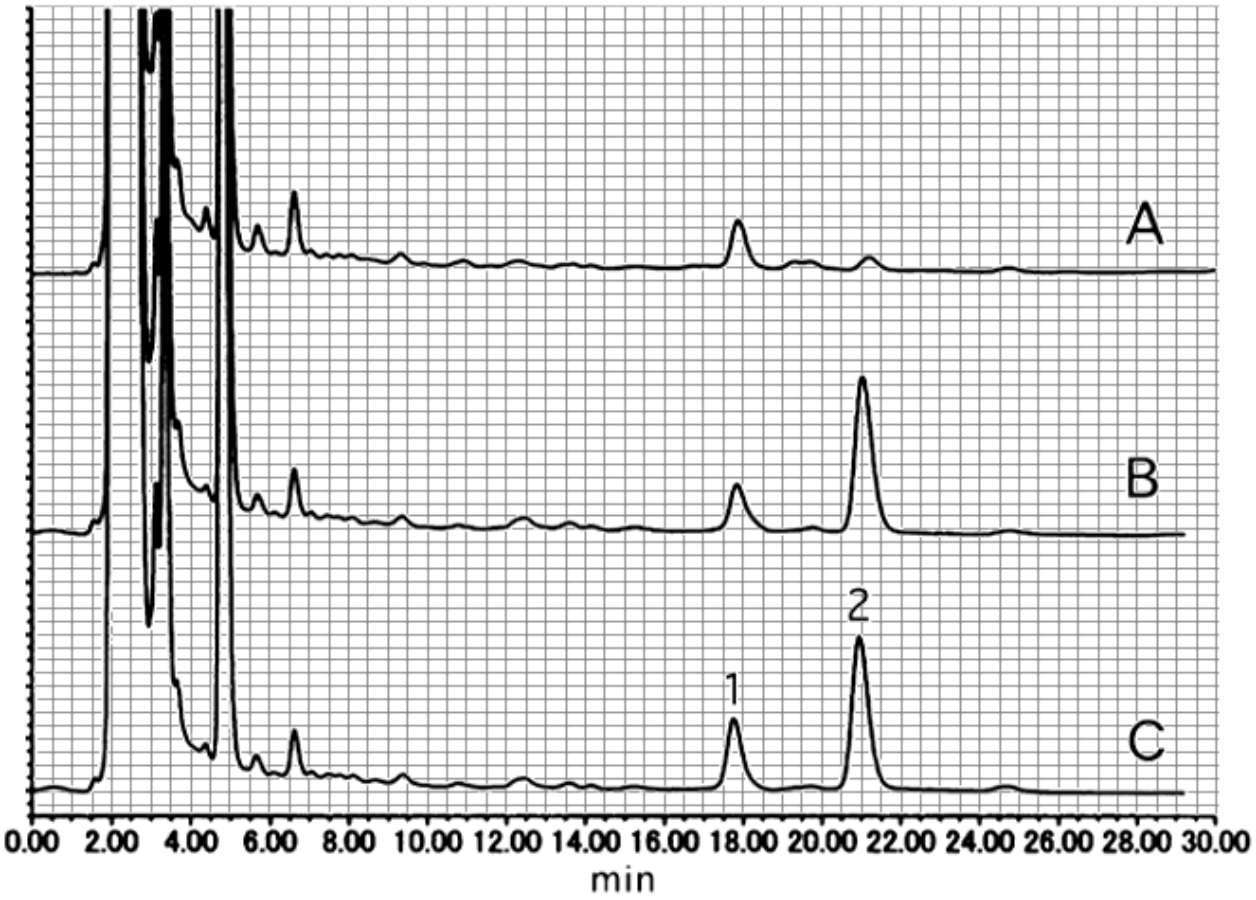
The blood sample of an influenza patient who ingested Tamiflu® was analysed using HPLC. An internal standard, mycophenolic acid, was used in the HPLC analysis and the mobile phase consisted of the potassium dihydrogen phosphate buffer at pH 3.0. The structures and pK_a values of mycophenolic acid and oseltamivir carboxylate, as well as the HPLC chromatograms are shown below.



mycophenolic acid
pK_a = 3.57



oseltamivir carboxylate
pK_{a1} = 4.13, pK_{a2} = 7.7



A: Blood sample without internal standard

B: Blood sample with internal standard (mycophenolic acid) added

C: Blood sample with internal standard (mycophenolic acid) and 1.0×10^{-6} g/ml oseltamivir carboxylate added

(e) **(i)** State whether normal-phase or reversed-phase HPLC was used in this analysis and account for the order of elution of the compounds.

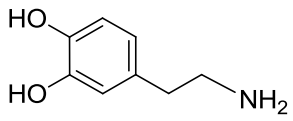
(ii) Predict how the retention time of oseltamivir carboxylate and mycophenolic acid will change if the mobile phase is buffered at pH 5.0 instead. Explain your answer.

(iii) Using the HPLC chromatograms B and C, determine the concentration (in g/ml) of oseltamivir carboxylate present in the blood sample.

[6]

[Total: 20]

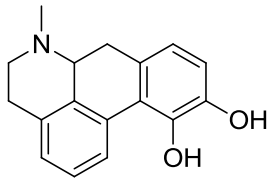
5 Dopamine is one of the principal neurotransmitters involved with stimulant activity in the brain. As such, drugs which can regulate the levels and concentrations of dopamine can lead to stimulant activity in the brain. Dopamine has the following structure:



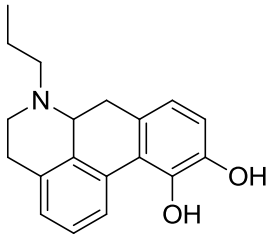
dopamine

(a) Dopamine levels can be controlled by the use of dopamine agonists and dopamine antagonists. Four particular drugs used in the regulation of dopamine levels are shown below:

agonist:

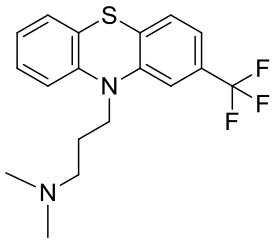


apomorphine

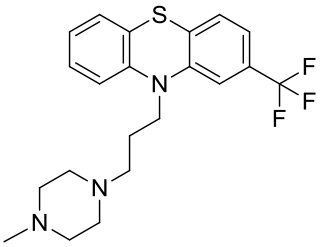


propylnorapomorphine

antagonist:



trifluoperazine



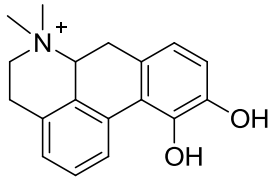
trifluoperazine

(i) Explain the meaning of the terms *agonist* and *antagonist*.

(ii) Draw the structure of apomorphine at physiological pH 7.4 and circle the pharmacophore that allows it to act as an agonist to the dopamine receptors.

(iii) By studying the structures of triflupromazine and trifluoperazine, suggest which features appear to be essential for them to act as an antagonist to dopamine receptors.

(iv) The above four drugs are designed to act on the dopamine receptors in the brain. However, the following derivative of apomorphine shows no stimulant activity even when introduced intravenously. Explain why.

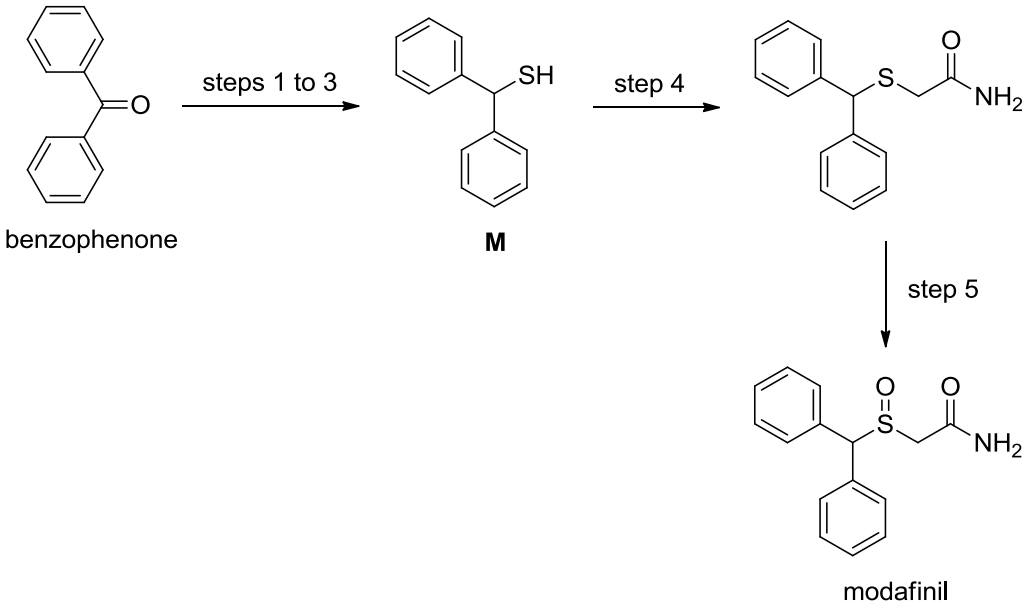


apomorphine derivative

[5]

(b) Modafinil, a type of dopamine re-uptake inhibitor, is a wakefulness-promoting agent used mainly in the treatment of disorders such as narcolepsy. However, the drug has also seen widespread off-label use such as cognitive enhancement. Due to the purported effect, it has been reported to be abused for performance enhancement during examinations.

One method of synthesis of modafinil is shown below:



benzophenone **M** modafinil

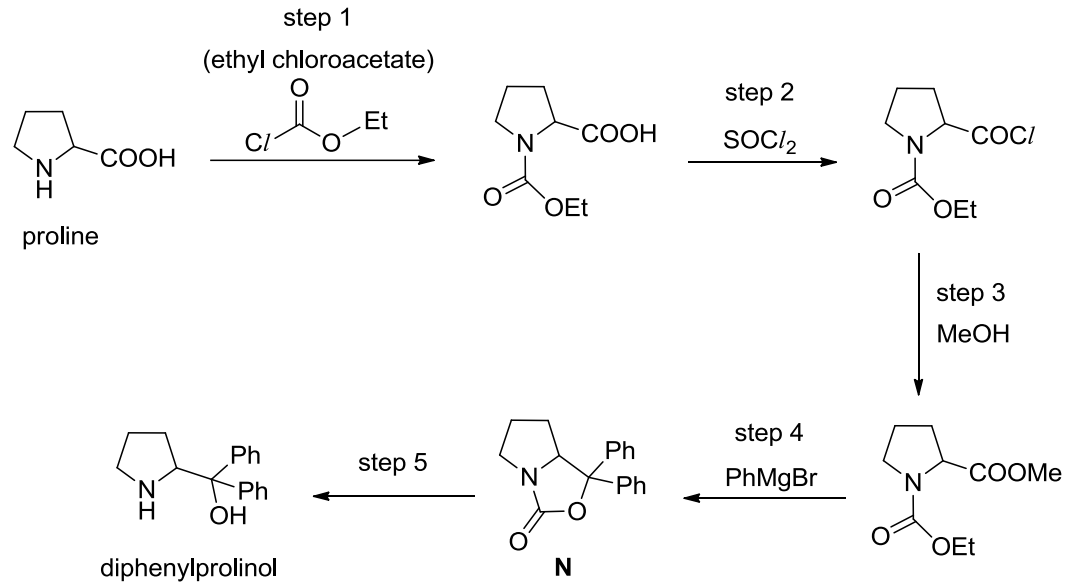
(i) Propose a 3-step synthesis of compound **M** from benzophenone. State clearly the reagents and conditions required, and draw the intermediate produced, in each step.

(ii) Suggest the reagents and conditions for steps 4 and 5.

(iii) This method of synthesising modafinil cannot produce an enantiomerically pure compound. Draw the structure of one of the enantiomers and assign the stereochemistry (*R* or *S*) at the chiral centre.

[7]

(c) Diphenylprolinol is also another type of dopamine re-uptake inhibitor. It is a designer drug which can be abused for recreational use to achieve feelings of euphoria and can be synthesised from the amino acid proline.



(i) Suggest the product formed if proline was reacted with $SOCl_2$. Hence, explain the purpose of using ethyl chloroacetate in step 1.

In step 4, the PhMgBr used is a Grignard reagent.

In Grignard reagents, the C–Mg bond is highly polar and the C atom becomes a nucleophilic centre. Hence, RMgBr behaves like $R^{-}\,{}^{+}MgBr$.

(ii) Propose the mechanism for step 4.

(iii) Suggest another product which may be formed in step 4.

(iv) State the reagent and conditions required for step 5.

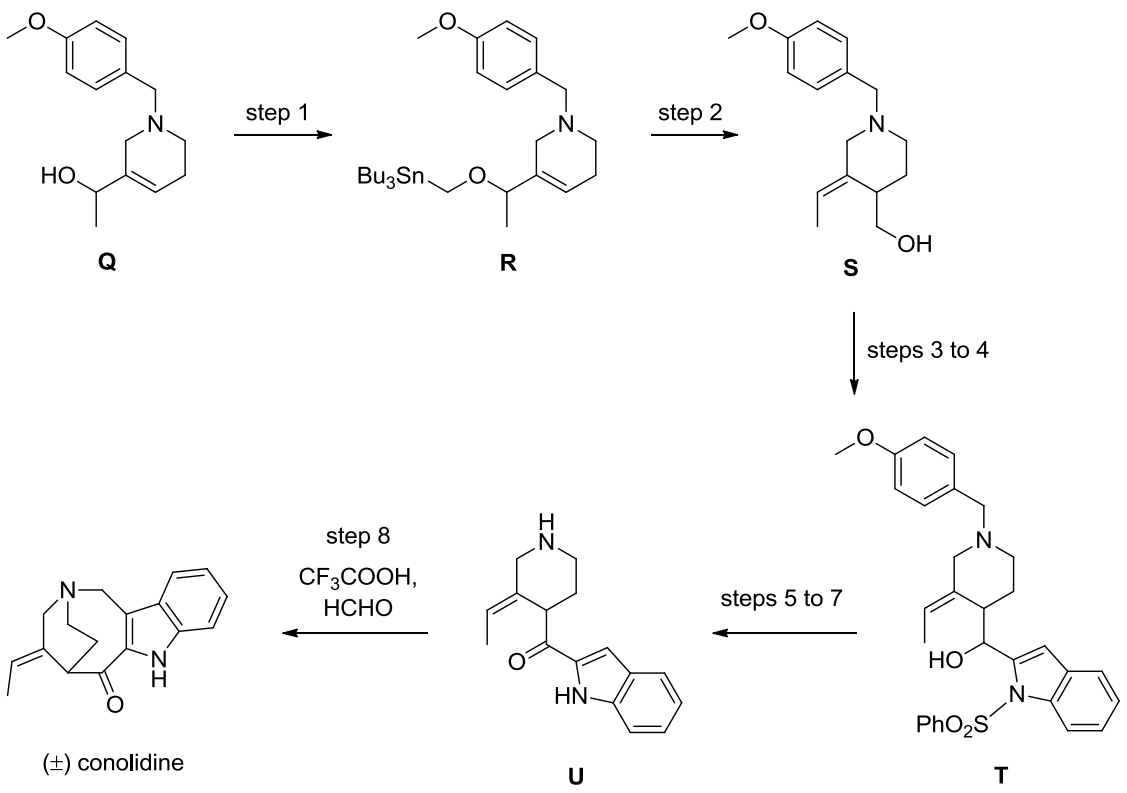
(v) $LiAlH_4$ is a source of hydride ions which act as nucleophiles during reduction reactions.

Draw the product formed when compound **N** is reacted with $LiAlH_4$ in dry ether.

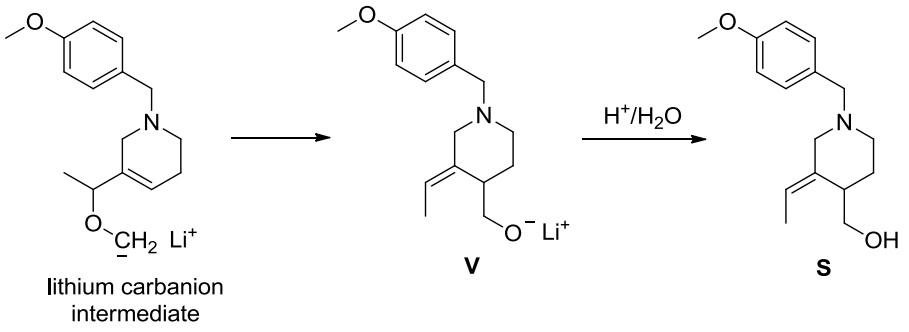
[8]

[Total: 20]

6 Conolidine is a rare plant-derived natural product believed to possess non-opioid analgesic properties. Its synthetic scheme is shown below.



(a) The conversion of **R** to **S** was achieved with the addition of butyllithium. In the reaction, a lithium carbanion intermediate was produced. The carbanion undergoes a concerted reaction to form **V**, which is then protonated to form **S**.



(i) Propose a concerted mechanism for the formation of **V** from the lithium carbanion intermediate. In your answer, you may ignore the stereochemistry of the alkene functional group in **V**.

(ii) Suggest why the above reaction occurs spontaneously.

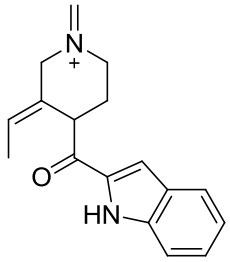
[2]

(b) Organolithium reagents (RLi) contain highly polar C–Li bonds and they behave like R^- Li^+.

The conversion of **S** to **T** involves two steps, one of which requires an organolithium reagent. Suggest reagents and conditions necessary for steps 3 and 4. [2]

(c) Assign the E/Z configuration of the alkene functional group in compound **U**. [1]

(d) In the conversion of compound **U** to conolidine, the following reaction intermediate **W** was found.



intermediate **W**

(i) Propose a mechanism for the conversion of intermediate **W** to conolidine.

(ii) Depending on the stereogenic configuration of **U**, either enantiomer of conolidine would be formed.

Draw the (*S*)-isomer of compound **U** and explain how it forms the corresponding enantiomer of conolidine. In your answer, you should show the stereochemistry of the enantiomer of conolidine formed.

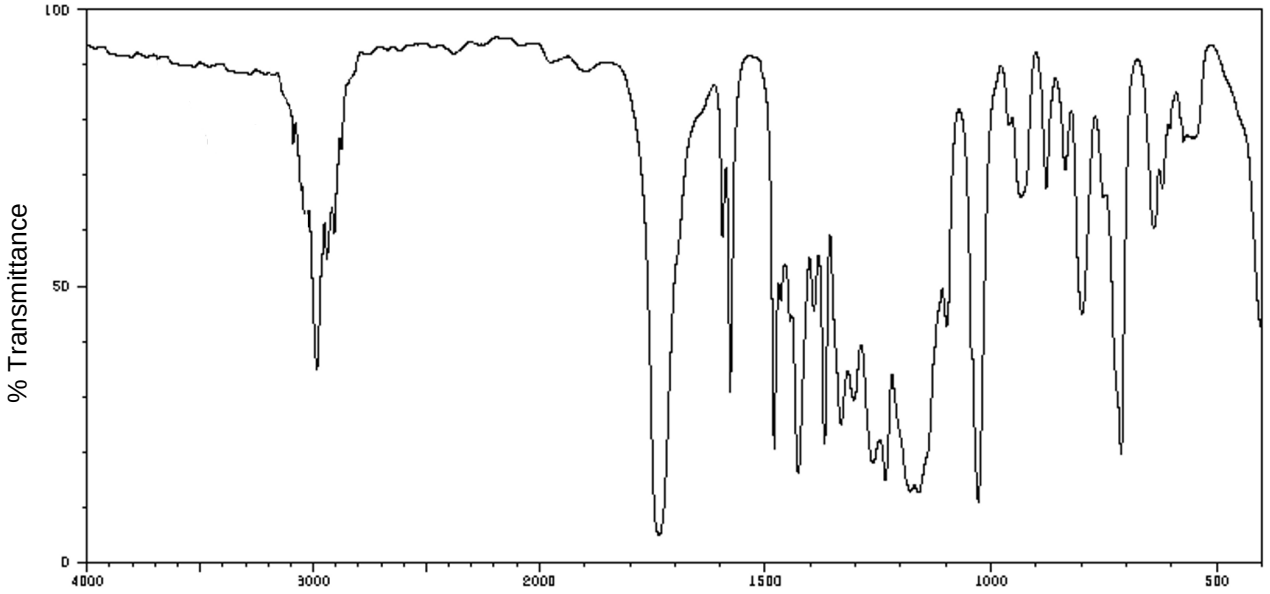
[4]

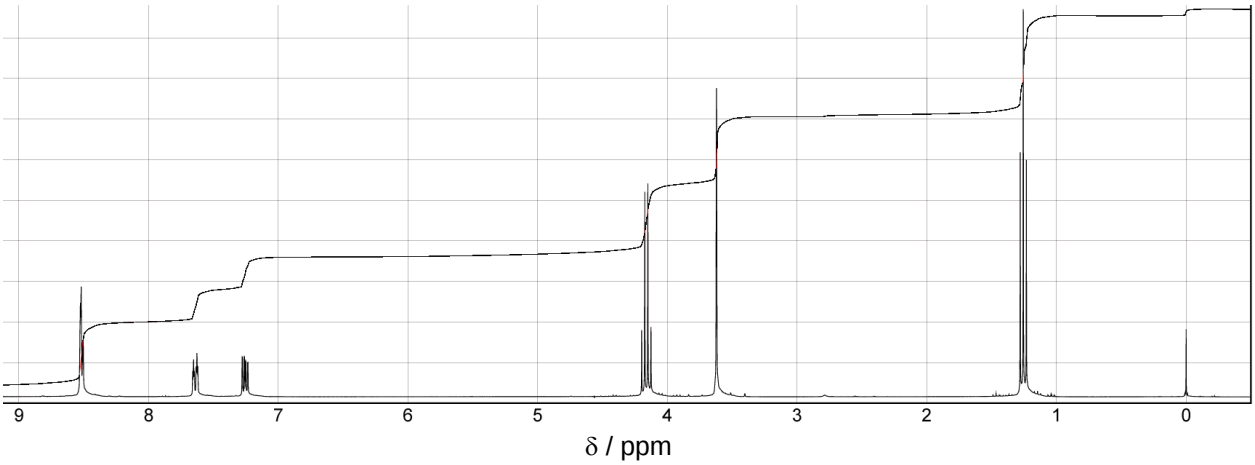
(e) Compound **Q**, the starting material in the synthesis of conolidine, can be produced from compound **P** via a multi-step synthesis.

Compound **P** contains carbon, hydrogen, oxygen and nitrogen only.

The mass spectrum, infra-red (IR) spectrum and NMR spectrum for compound **P** are provided below.

m/e	Relative abundance
29	53
92	100
165 (M^+)	39
166	4





(i) By determining the identities of the peaks in the mass spectrum and using the IR and NMR data, suggest the structure of compound **P**. Explain your reasoning.

(ii) In the preparation of samples for NMR analysis, samples are frequently dissolved in deuterated chloroform ($CDCl_3$). In these NMR spectra, a small singlet peak at δ 7.26 ppm can be typically seen.

Suggest how this peak arises.

[11]

[Total: 20]

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