CANDIDATE NAME | | CT GROUP | 16S

CENTRE NUMBER | | INDEX NUMBER |

PHARMACEUTICAL CHEMISTRY

9812/01

Paper 1

21 September 2017

2 hours 30 minutes

Additional Materials: Answer Paper
                                Data Booklet

READ THESE INSTRUCTIONS FIRST

Write your index number, name and CT on all the work you hand in.
Begin each question on a **fresh** sheet of writing paper. A **nil return** is necessary for any unattempted question.
Write in dark blue or black pen.
You may use a soft pencil for any diagrams or graphs.
Do not use staples, paper clips, glue or correction fluid.

Answer any **five** questions.
At the end of the examination, fasten all your work securely together.

The number of marks is given in brackets [ ] at the end of each question or part question.
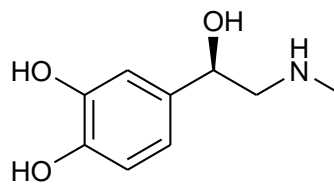The use of an approved scientific calculator is expected, where appropriate
You are reminded of the need for good English and clear presentation in your answers.

1 (–)Zilpaterol is a β-adrenergic *agonist*. β-agonists are known to cause heart palpitations and nausea. The natural agonist for the β-adrenergic receptor is adrenaline.
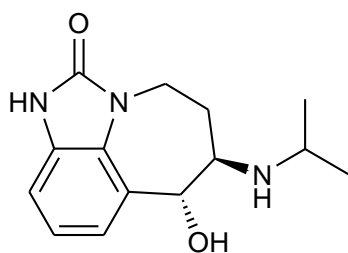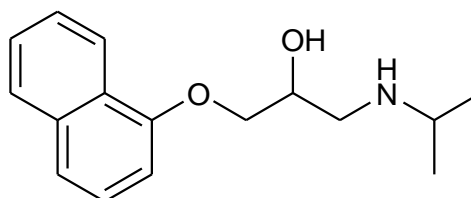

(–)Zilpaterol   Adrenaline

(a) **(i)** Explain the term '*agonist'*.   [1]

(ii) Suggest two possible types of interaction that allow adrenaline and zilpaterol to bind with the β-adrenergic receptor.   [2]

(iii) β-blockers, such as propranolol, are used to block the adverse effects caused by adrenaline.


Propranolol

Suggest the role that propranolol plays with respect to the β-adrenergic receptor and explain your answer.   [2]

(b) The concentration of (–)zilpaterol and adrenaline in meat samples may be determined by chromatography. The meat sample is first extracted using an aqueous acidic solution. Measured volumes of the extract are analysed using reverse-phase HPLC with methanol-water mobile phase.

(i) Explain why GLC and paper chromatography are less suitable methods of analysis of the concentrations of (–)zilpaterol and adrenaline as compared to reverse-phase HPLC.   [2]

(ii) Suggest how you would distinguish (–)zilpaterol from other compounds as it emerges from the column and how you would measure its concentration.   [2]

(iii) Predict which molecule, (–)zilpaterol or adrenaline, will be eluted from the column first.   [1]

(c) The first part of the synthesis of zilpaterol is shown in the reaction scheme below.
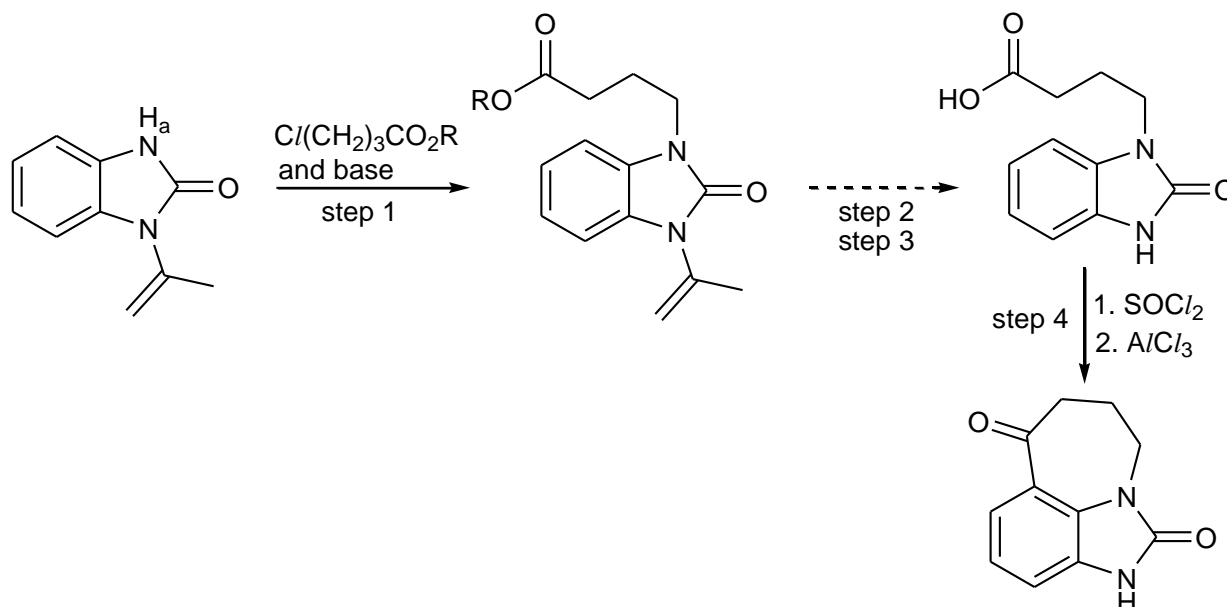
(i) Explain, using resonance structures, why H_a is acidic. [2]

(ii) Suggest the type of reaction taking place in step 1. [1]

**(iii)** Explain how $SOCl_2$ and $AlCl_3$ bring about reaction in step 4. [2]

(d) The product from step 4 is reacted with alkyl nitrite (RONO) and subsequently, zilpaterol is formed.
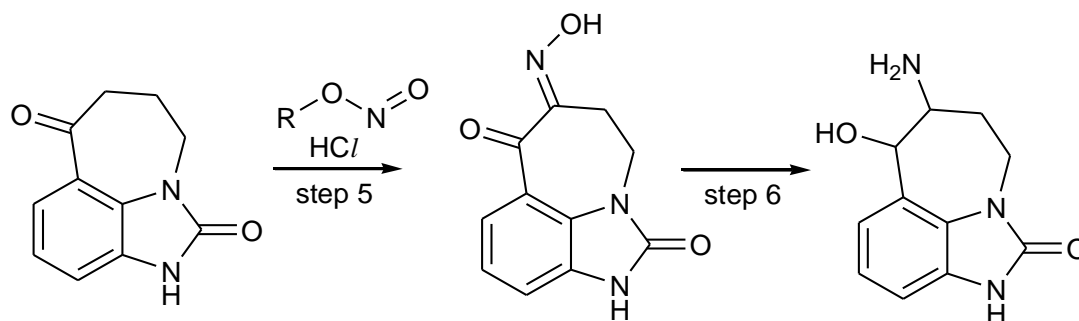

(i) Suggest a mechanism for step 5, given the following information:

- Carbonyl oxygen is first protonated by HCl and subsequently, an enol is formed.
- ROH is formed as a product. [4]

(ii) Suggest the reagents and conditions for step 6. [1]

[Total: 20]

2 **(a)** The penicillins belong to a class of antibacterials which function by inhibiting cell wall synthesis in bacteria. They inhibit the transpeptidase enzyme by mimicking the action of its natural substrate, the acyl-D-Ala-D-Ala portion of the polysaccharide chain in bacteria cell wall.
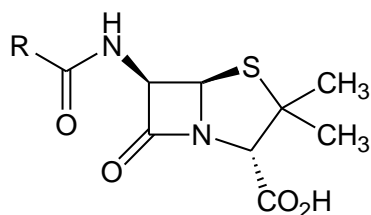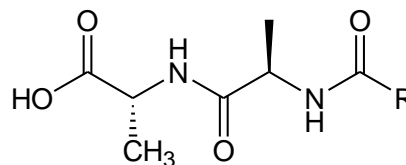

the penicillins   acyl-D-Ala-D-Ala

(i) State two other ways in which antibacterials work. [2]

(ii) Redraw the structure of the acyl-D-Ala-D-Ala in an orientation to show how penicillins are able to mimic its action. [1]

(b) The β-lactam ring in penicillins is easily hydrolysed in an acidic medium. This is made easier by the acyl side chain. The first step of the mechanism is shown below. An intermediate **X** is formed which then further hydrolyses to form a penicilloic acid **Y**.
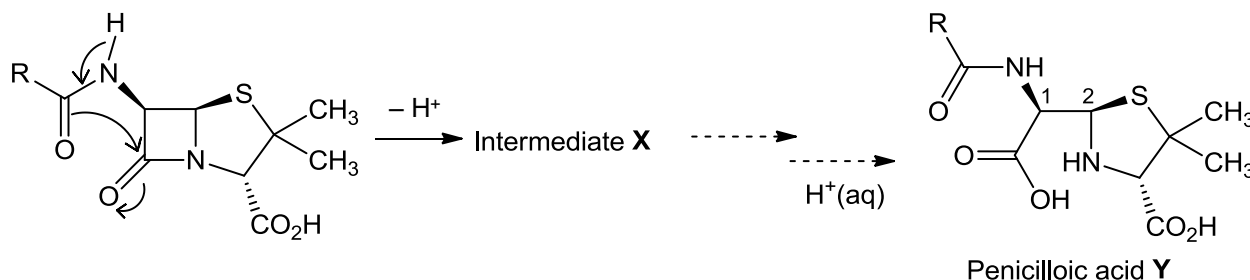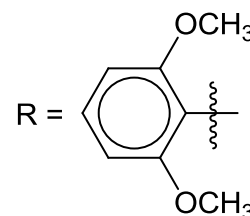

Penicilloic acid **Y**

(i) Draw the structure of intermediate **X**. [1]

(ii) Complete the mechanism to show how penicilloic acid **Y** is formed from **X**. [3]

(iii) Assign R/S configurations to all chiral carbons in **Y**. [1]

(iv) Draw Newman projections along C1-C2 to show an eclipsed and a staggered conformation of **Y**. Label your answers clearly. [2]

(v) The structure of the R group in three penicillins are given below.
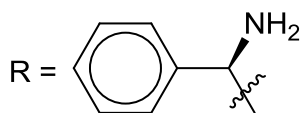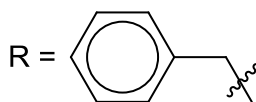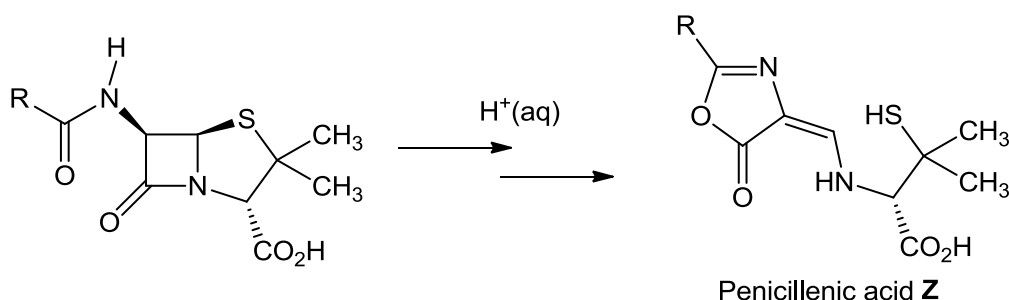

ampicillin   penicillin G   methicillin

Rank these three penicillins in order of increasing stability in an acidic medium. Explain your answer. [3]

(c) In the acid-catalysed degradation of penicillins, penicillenic acid **Z** can be formed as a side product. This forms the basis for the determination of penicillins via UV spectroscopy.

Penicillenic acid **Z**

The graph below shows the UV absorption curves for a 0.025 mg cm^{-3} sample of sodium penicillin G ($C_{16}H_{17}N_2NaO_4S$) before and after heating in a pH 4.6 acetate buffer for 15 min. The absorbance values were measured from 240 nm to 400 nm using a cell of path length 1.0 cm.

- - - - - Before heating ——— After heating
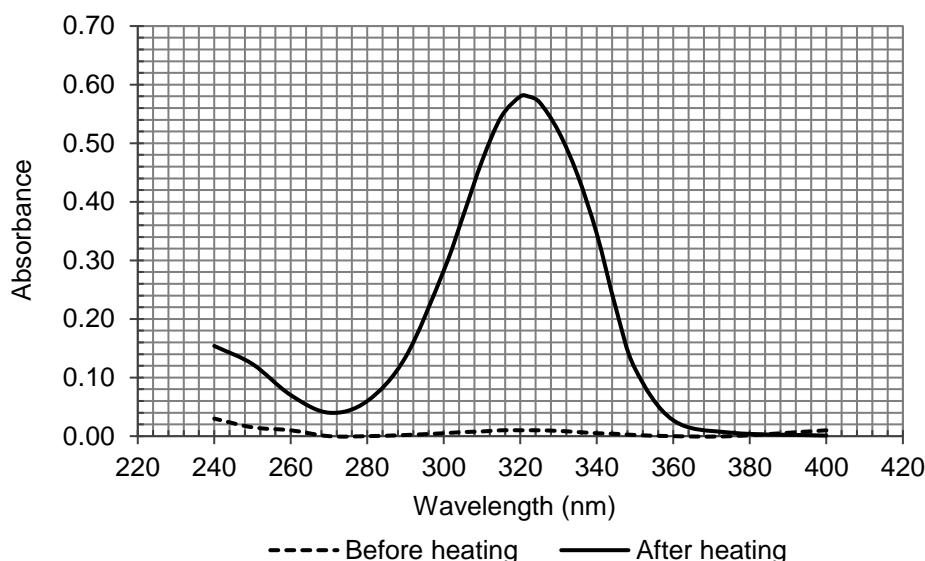
(i) By considering the molecular structures of the species present, explain why a significant absorption is observed for the sample after heating. [1]

(ii) Given that the molar mass of the absorbing species is the same as that for sodium penicillin G, use the graph above to calculate the molar extinction coefficient of the absorbing species at 322 nm. [2]

(d) A second method of analysis of penicillins involves the use of copper(II) acetate which reacts in a 2:1 ratio with the penicillin at pH 6 to 6.8 to give a green complex.

In an experiment involving penicillin G (M_r = 334.1), the green complex had an absorption maximum at 650 nm with molar extinction coefficient of 240 mol^{-1} dm^3 cm^{-1}. Beer's Law was found to be valid over the concentration range of 20 to 1000 μg cm^{-3} of penicillin G.

A commercial solution of penicillin G was labelled as containing approximately 150 mg of penicillin G in 100 cm^3. A 20.0 cm^3 sample of this solution was diluted to 100 cm^3 with water. The diluted solution gave an absorbance of 0.230 at 650 nm in a cell with path length of 1.0 cm.

(i) Calculate the actual mass of penicillin G in 100 cm^3 of the commercial solution. [3]

(ii) Explain why it was necessary to dilute the commercial solution of penicillin G before analysis. [1]

[Total: 20]

3 (a) Benzodiazepines (BZs) have been widely used therapeutically for their ability to reduce anxiety and act as tranquilizers by enhancing the effect of GABA (γ–aminobutyric acid) through binding to the GABA$_A$ receptor. The generic structure of BZs is shown below.
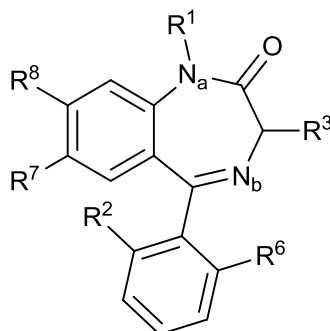
A series of BZ analogues were synthesised and a study was conducted to examine the effects of different substituents R^1, R^6 and R^7 of BZ. R^2, R^3 and R^8 were unmodified in this study and you may assume that they are hydrogen atoms. Three main factors were identified to affect the binding affinity of the BZ to the BZ binding site:

- **Aromatic group dipole** (μ), which can be taken to be the net dipole moment of a monosubstituted benzene ring
- **Lipophilicity** (π), which can be interpreted as how non-polar a substituent is
- **Molar refractivity** (MR), which can be roughly taken to be a measure of the volume occupied by an atom/group of atoms

The binding affinity can be measured using the physical quantity lg IC$_{50}$. <u>The smaller the value of lg IC$_{50}$, the better the binding of the drug to the binding site</u>.

Table 3.1

Substituent	Magnitude of aromatic group dipole (μ)	Lipophilicity (π)*	Molar refractivity (MR)
−H	0.00	0.00	1.03
−CH$_3$	0.36	0.56	5.65
−Cl	1.59	0.71	6.03
−OH	1.59	−0.67	2.85
−NHOH	0.14	−1.34	7.22

Note: *a negative value denotes low lipophilicity

Table 3.2

Analogue	R_7	R_1	R_6	lg IC$_{50}$
A	H	CH$_3$	H	1.15
B	CH$_3$	CH$_3$	H	0.71
C	NHOH	CH$_3$	H	1.98
D	NO$_2$	H	H	0.97
E	NO$_2$	CH$_3$	H	0.78
F	NO$_2$	OH	H	0.43
G	Cl	H	H	0.26
H	Cl	H	Cl	0.85
I	Cl	H	NHOH	1.27

(i) Based on the information provided, suggest which of the three factors, μ, π or *MR*, has the greatest impact on the binding affinity of the drug for:

    I: R^7
    II: R^1
    III: R^6

You should state clearly what the effect of the stated factor is on the binding of the drug to the binding site, with supporting evidence from the data given. [6]

(ii) When analogue **A** was modified in a separate experiment such that a chlorine atom takes the position of R^3, a mixture of two enantiomers was obtained. Suggest a method to separate the two enantiomers. [1]

(iii) It is postulated that by modifying the substituent R^2, the electron density on N_b can be greatly increased and improve the binding of the BZ to the BZ binding site greatly.

Copy the structure of the generic BZ on your answer paper and replace R_2 with a suitable substituent from **Table 3.1**. Draw appropriate structures to illustrate how your substituent can greatly increase the electron density on N_b. [2]

(iv) Based on your answer in **(a)(i)** and information given in **(a)(iii)**, suggest **two** features that might be present in the BZ binding site. Your answer should also include any possible interactions that the BZ might have with the BZ binding site. [2]

(b) Chlordiazepoxide is an example of a benzodiazepine used to treat anxiety. Like other BZs, its mode of action involves binding to the $GABA_A$ receptor to enhance the effect of GABA. Chronic use of chlordiazepoxide can lead to the development of *tolerance* to the drug.

(i) Explain how *tolerance* to chlordiazepoxide arises in the body. [2]

Chlordiazepoxide can be synthesised as shown.

chlordiazepoxide
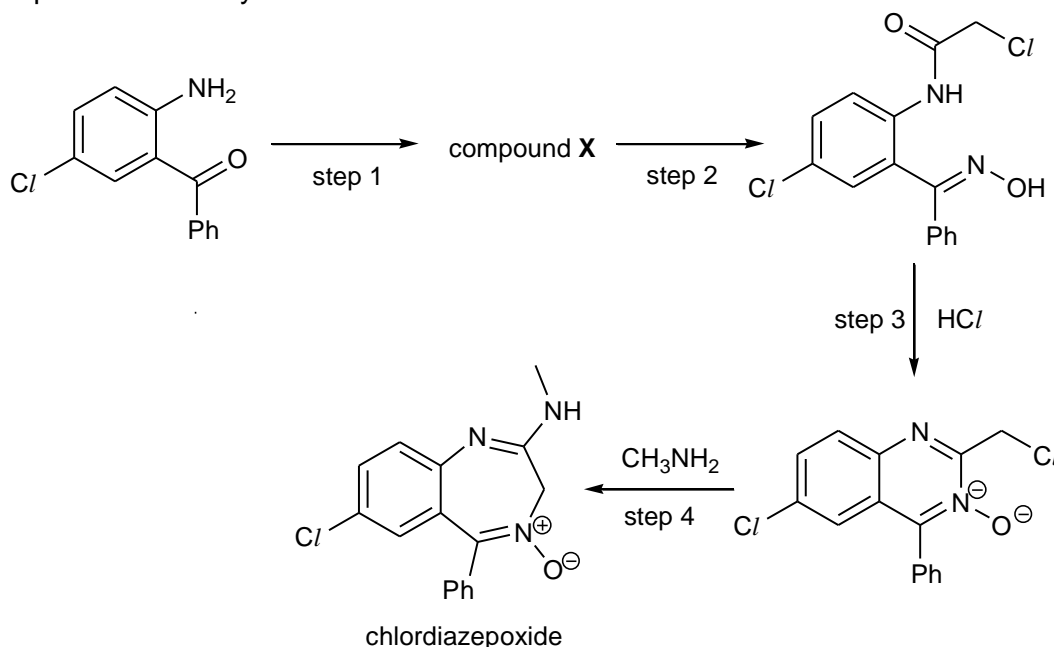
(ii) Suggest reagents for step 1 and step 2 and draw the structure for compound **X.** [3]

(iii) Draw the mechanism for step 3. [3]

(iv) Draw a possible side product that could be formed in step 4. [1]

[Total: 20]

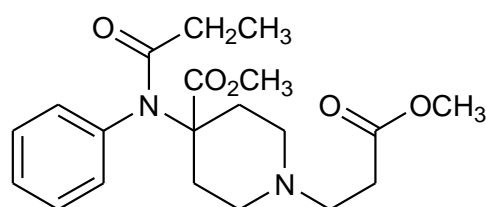**4 (a)** Read the passage below and answer the questions that follow.

On 23 October 2002, Chechen militants seized the Dubrovka Theatre in Moscow during a performance, holding 900 people hostage. 56 hours later, following failed negotiations, a "knockout gas" was administered into the theatre's ventilation system. Within minutes, the theatre occupants were overcome by the gas, enabling Russian troops to storm the theatre, kill all militants and rescue the hostages. However, many victims were unconscious. Owing to a lack of information provided to medical emergency responders, who anticipated treating victims of gunfire instead of chemical agents, the lives of more than 130 hostages were lost, many to suffocation due to respiratory depression. Many who had been saved had been administered naloxone, a known antidote to opioid poisoning.

Interviewed after awakening in the hospital, a victim recalled vomiting and seeing blood, but strangely felt no pain in his stomach. Another casualty drifted in and out of consciousness and was surprised to feel no pain when a drip was inserted into her arm. Under international pressure, the Russian government revealed that the chemical was a fentanyl derivative. Upon analysis of urine and clothing from victims, a British team later concluded that remifentanil was used in the "knockout gas".
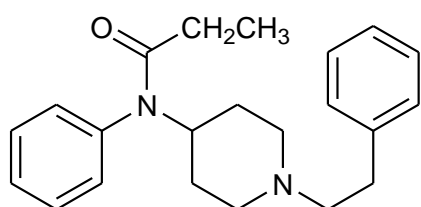
fentanyl
pK_a of (protonated) ring nitrogen = 8.4

remifentanil
pK_a of (protonated) ring nitrogen = 7.1

(i) What evidence suggests that the "knockout gas" contained an *analgesic*? [1]

(ii) Discuss whether the analgesic used was likely to be *narcotic* or *non-narcotic*. [1]

(iii) The drug used needed to have a fast onset of action (so that militants will be quickly overcome) and a short duration of action (so that hostages may recover quickly). Therefore, it needed to exhibit the following characteristics:

- Ease of reaching the target
- Ease of metabolism into inactive metabolites that could be easily excreted

Given that the physiological pH is 7.4, calculate the [deprotonated form]/ [protonated form] ratio of both fentanyl and remifentanil and suggest why remifentanil was preferred over fentanyl under both criteria. [3]

(iv) Would naloxone be an opioid receptor agonist or antagonist? Explain. [1]

(b) By studying their molecular structures, state **one** difference in the IR spectra of fentanyl and remifentanil. Identify the wavenumbers of the absorptions and the functional group involved. [1]

(c) Sufentanil ($C_{22}H_{30}N_2O_2S$) is another synthetic opioid analgesic drug that is 5 times more potent than fentanyl. It is **structurally similar** to remifentanil ($C_{20}H_{28}N_2O_5$) and their difference lies in the two methyl ester groups in remifentanil being replaced with other groups.

The ^{1}H NMR spectrum of Sufentanil is shown in the table below.

proton chemical shift value, δ	splitting	number of protons
1.14	triplet	3
1.70	triplet	4
2.27	quartet	2
2.29	triplet	4
2.65	triplet	2
2.69	triplet	2
3.24	singlet	3
3.44	singlet	2
6.74 – 7.06	multiplet	3
7.10 – 7.31	multiplet	5

Deduce the structure of sufentanil. Explain your reasoning concisely. You need not assign all the peaks. [3]

(d) Allicin is a compound found in crushed garlic. Diallyl disulfide is a principal component of the distilled oil of garlic and it is a precursor to allicin in a typical laboratory synthesis.


allicin   diallyl disulfide
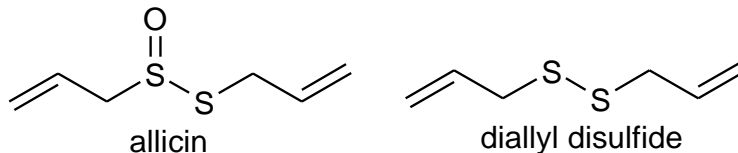
(i) Suggest a synthesis of diallyl disulfide from 3-bromopropene. You should give all reagents and conditions and intermediate compounds. [2]

(ii) Diallyl disulfide undergoes oxidation with hydrogen peroxide to give allicin. Write out a half equation for the oxidation of diallyl disulfide to allicin in acidic conditions and hence write a balanced equation for the oxidation of diallyl disulfide using hydrogen peroxide. [2]

(iii) Under suitable conditions, one molecule of allicin decomposes into two organosulfur molecules, thioacrolein and compound **A**. Two molecules of compound **A** may undergo a condensation reaction to give one molecule of allicin.
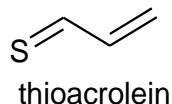
thioacrolein

Suggest the structure of compound **A**. [1]

(iv) Thioacrolein readily takes part in the Diels-Alder reaction, a useful method linking two molecules with the formation of a new ring in one single step. The formation of 3,4-dihydro-3-vinyl-1,2-dithiine is shown below.

3,4-dihydro-3-vinyl-1,2-dithiine
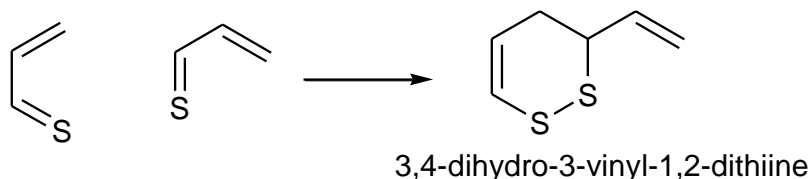
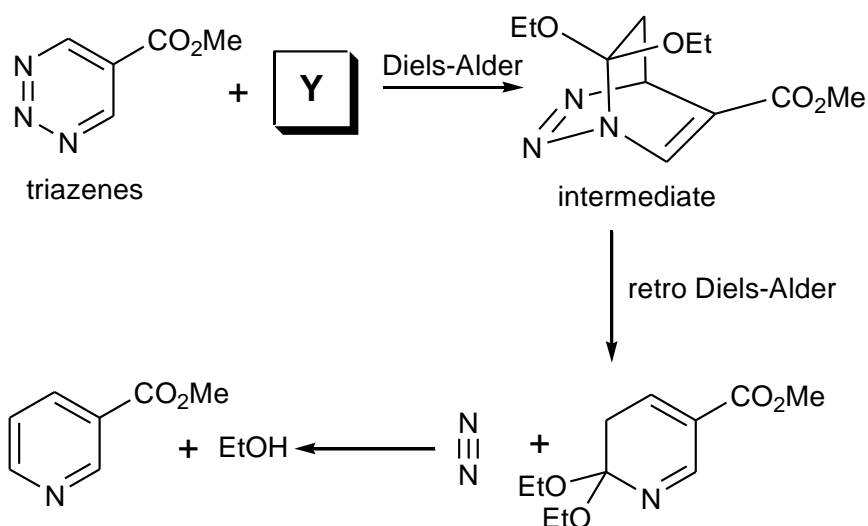Use curly arrows to suggest a mechanism for this reaction. [1]

(v) The structure of the other isomer formed from the Diels-Alder reaction is 2,4-dihydro-2-vinyl-1,3-dithiine. Using the name of the compound given in **(d)(iv)** as a clue, suggest its structure. [1]

(e) It is possible for the product in the reaction described in **(d)(iv)** to undergo a retro Diels-Alder reaction to give back the starting material under suitable conditions.

Pyridines are important moieties in natural product. Triazenes are able to form pyridines via two key processes – a Diels-Alder reaction followed by a retro Diels-Alder reaction.

triazenes   +   **Y**   Diels-Alder   intermediate

retro Diels-Alder

CO_2Me + EtOH ←   + CO_2Me

(i) Suggest a structure for **Y**. [1]

(ii) Use curly arrows to suggest a mechanism for the retro Diels-Alder reaction. [1]

(iii) Suggest the driving force for the retro Diels-Alder reactions as well as the step leading to the pyridinium product. [1]

[Total: 20]

5 **(a)** There are two main routes by which nucleophilic substitution reactions take place, the S_N1 and the S_N2 mechanisms.

The table below summarises the relative rates of S_N1 (denoted by k_1) and S_N2 (denoted by k_2) reactions of 4 simple alkyl bromides.

Alkyl bromide	CH_3Br	CH_3CH_2Br	$(CH_3)_2CHBr$	$(CH_3)_3CBr$
Relative k_1	2×10^{-2}	4×10^{-2}	1	4×10^6
Relative k_2	6×10^3	30	1	5×10^{-5}

The experiment conducted to measure k_1 uses HCO_2H (as both the nucleophile and solvent) at 100 °C. The experiment conducted to measure k_2 uses radioactive $^{82}Br^-$ (as the nucleophile) and propanone (as the solvent) at 25 °C.

(i) Using the information above, describe and explain the conditions that would favour S_N2 mechanism over the S_N1 mechanism. Your answers should cover the following:

- The features of the substrate
- The nature of the solvent
- The nature of the nucleophile [3]

(ii) Suggest reasons why the following reactions occur in the way shown.

($-CH_2CH_3$ is represented as $-Et$)
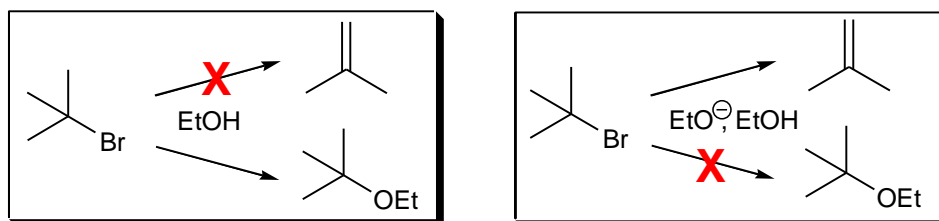
[2]

It is often difficult to distinguish between an E1 mechanism from an E2 mechanism. A useful tool used in the mechanism study is to replace hydrogen with deuterium (^2H), giving rise to what is commonly known as the kinetic isotope effect.

A snapshot of the energy profile diagram showing the minimum amount of energy required to homolytically cleave the C–H and C–D bond is shown in Figure 5.1.

The zero point energy, E_o can be found by the following equation:
$$E_o = \tfrac{1}{2}hc\nu \text{ (unit: J)}$$

where 'h' is the planck constant, 'c' is the speed of light and 'ν' is the wavenumber.
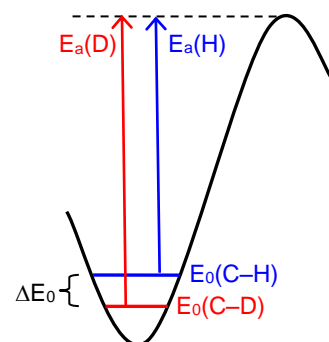

Figure 5.1

(iii) Using the information above and the *Data Booklet*, show that $\Delta E_o \approx 4.8$ kJ mol^{-1}. The C–H and C–D stretching vibrations are 3000 cm^{-1} and 2200 cm^{-1} respectively. [2]

**(iv)** By using the Arrhenius equation, $k = Ae^{(-Ea/RT)}$, show that the rate of homolytic cleavage of a C–H bond is seven times as fast as that of a C–D bond at 25 °C. Assume that the pre-exponential factor, A, for both cleavages are identical. [1]

(v) Hence suggest how the use of deuterium labelling allows chemists to decipher whether E1 mechanism or E2 mechanism occurs when $(CH_3)_3CBr$ is subjected to EtO$^-$ in EtOH. [2]

(b) Selexipag and its active metabolite, the corresponding carboxylic acid, are non-prostanoid PGI-2 receptor agonists. Its synthesis is shown below.
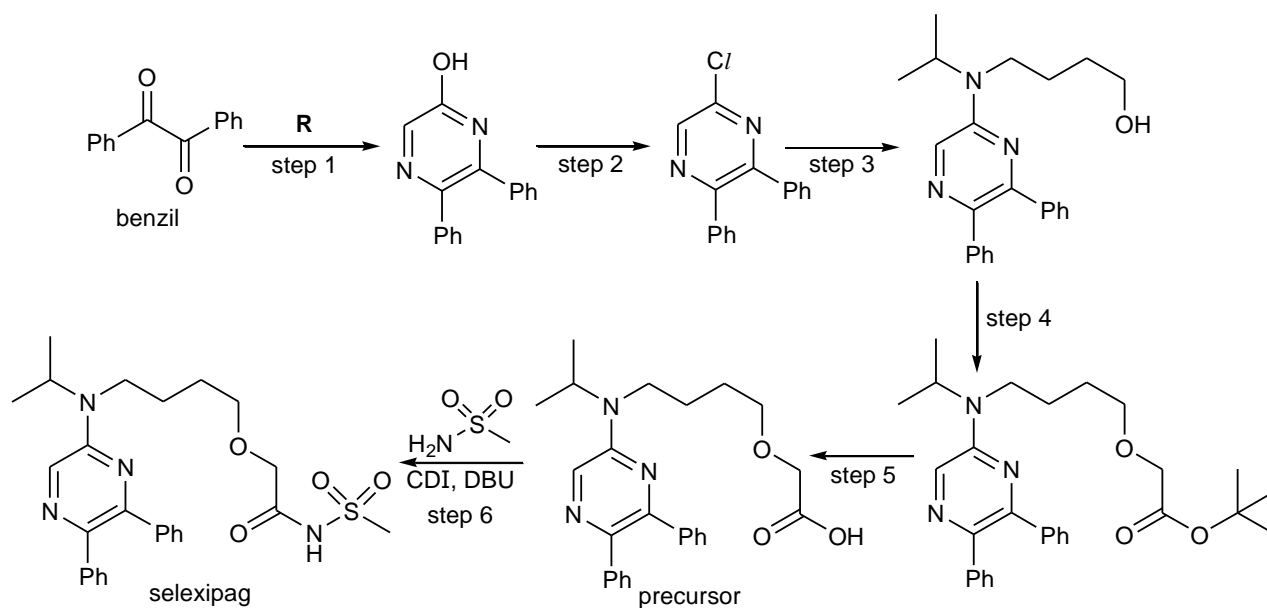

(i) Given that for step 1, one equivalent of **R** undergoes condensation reaction with benzil to generate two equivalents of water, suggest the identity of **R**. [1]

(ii) Suggest reagents for step 3 and step 5. What types of reactions are involved in step 3 and step 5? [2]

(iii) In step 6, CDI is used as the "coupling agent" for the formation of the amide-like linkage in selexipag and DBU is used as the base. The structures of CDI and DBU, together with a key intermediate are given as follow.
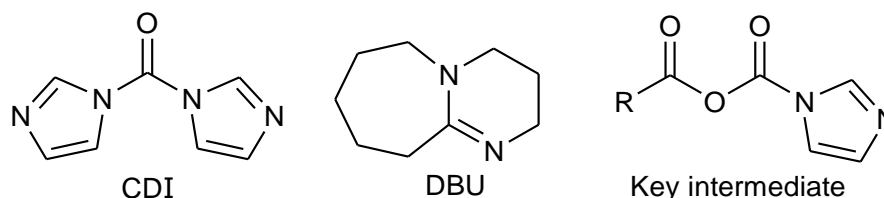
Step 6 begins with an initial deprotonation of the precursor by DBU to generate a carboxylate anion. This is followed by a reaction with CDI to generate the key intermediate. Finally the key intermediate reacts with $CH_3SO_2NH_2$ giving selexipag with the expulsion of gas **S**.

By representing the precursor as RCO_2H, suggest the mechanism to step 6 and hence identify gas **S**. [4]
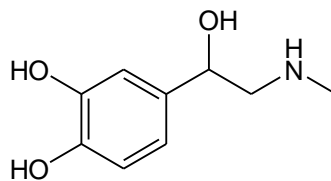
(c) Selexipag contains a pyrazine ring.
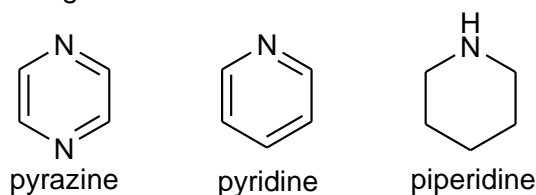
Arrange the three compounds above in order of increasing pK_b and explain the difference in their basicity. [3]

[Total: 20]

6 (a) Adrenaline is a naturally produced hormone that can act as a stimulant by binding to the adrenergic receptors.
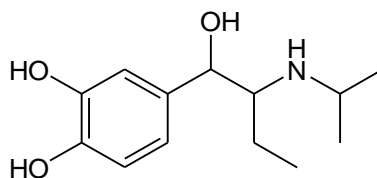

adrenaline

(i) What is meant by the term *stimulant*? [1]

(ii) Compare and contrast adrenaline and amphetamines, in terms of their chemical structures and physiological effects. [2]

(b) In the treatment of asthma, a specific type of receptor known as β_2–adrenergic receptors in the bronchial smooth muscles has to be stimulated. This leads to the dilation of the airways in the lungs. Even though adrenaline can be used for the treatment of asthma, it binds with all available adrenergic receptors in the body, causing unwanted side effects. Isoprenaline, isoetharine and salbutamol are β_2–adrenergic receptor agonist that are used to replace adrenaline in the treatment of asthma.
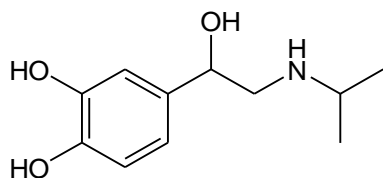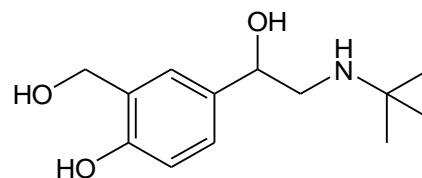
isoprenaline   isoetharine   salbutamol

A ^{1}H NMR spectrum of isoprenaline ($C_{11}H_{17}NO_3$) in the presence of D_2O is shown in the table below.

proton chemical shift value, δ	splitting	number of protons
1.37	doublet	6
3.10	doublet	2
3.58	multiplet	1
4.84	triplet	1
6.76 – 6.91	multiplet	3

(i) Identify the protons responsible for the resonances at δ 1.37, 3.10, 3.58 and 4.84 and account for the splitting observed. [2]

Besides ^{1}H NMR, the determination of the structure of an unknown organic compound can also be made through ^{13}C NMR spectroscopy. ^{13}C NMR spectra provides direct information about the carbon skeleton by giving us a glimpse into the number of non-equivalent carbons. Most of the principles behind ^{1}H NMR apply to the study of ^{13}C NMR. One key difference is that ^{13}C NMR spectrum are often obtained without coupling, i.e. each C signal appears as a singlet.

Two types of ^{13}C NMR spectra of isoprenaline are shown on the next page. Figure 6.1 shows a typical ^{13}C spectrum; Figure 6.2 shows a ^{13}C spectrum obtained through a technique known as DEPT-135 (Distortionless Enhancement by Polarisation Transfer) where all the C with an odd number of H give a positive peak and all the C with an even number of H give a negative peak. Those C without H give no peak.
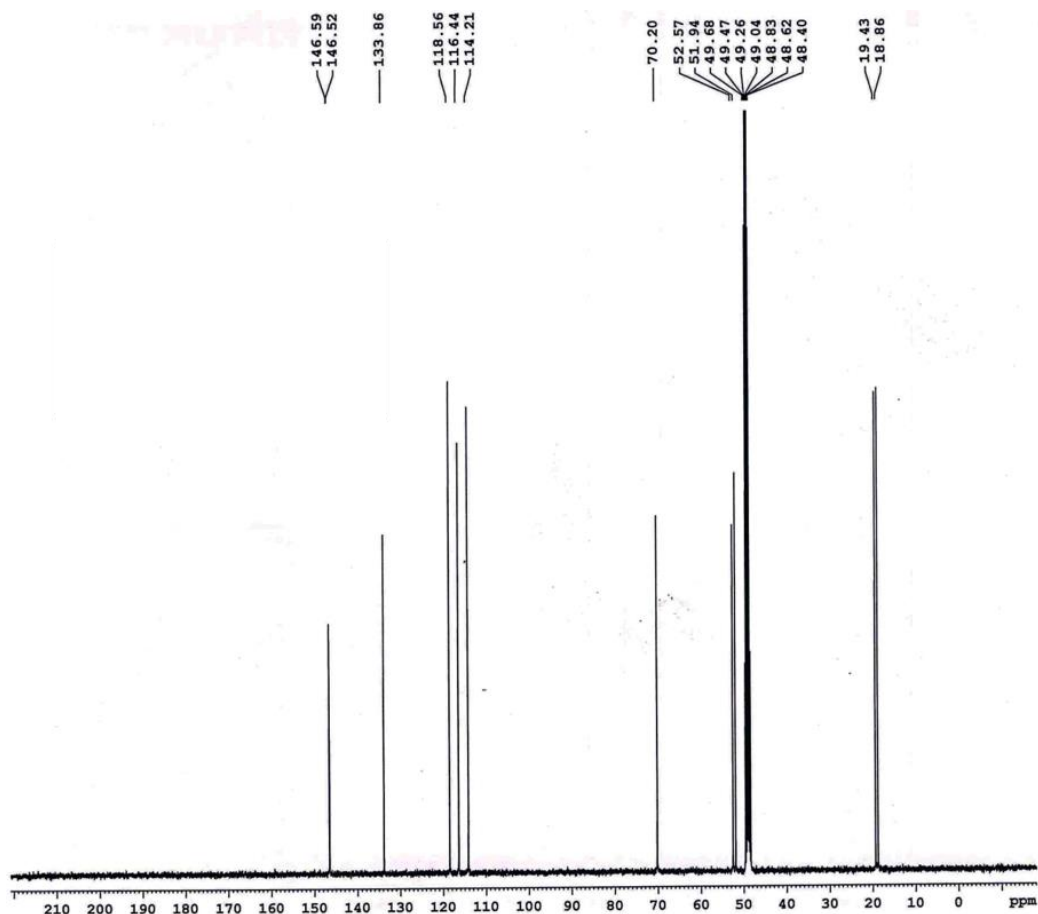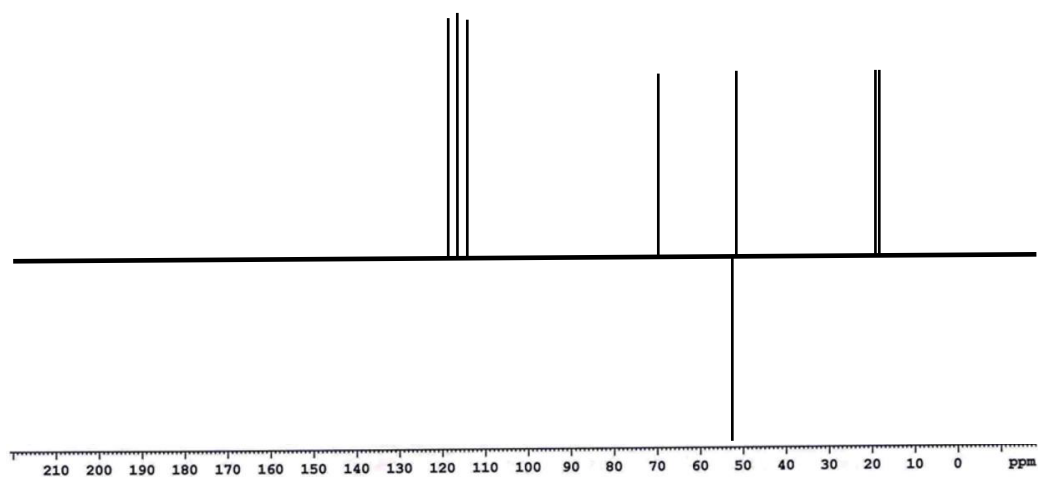
146.59
146.52
133.86
118.56
116.44
114.21
70.20
52.57
51.94
49.68
49.47
49.26
49.04
48.83
48.62
48.40
19.43
18.86

210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 ppm

Figure 6.1

210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 ppm

Figure 6.2

(ii) Using the two spectra above, assign the carbons responsible for all the signals besides the signals from 48.40 to 49.68 ppm as they are due to the solvent. [2]

(iii) By studying the structures of isoprenaline, isotharine and salbutamol, describe how you would use ^{1}H and/ or ^{13}C NMR to identify the three drugs.

In your answer, you should identify unique chemical shift values and their corresponding splitting pattern. [2]

(c) Compound **A** (M_r = 272.9) which contains the elements of carbon, hydrogen, oxygen and a halogen, is an intermediate in the synthesis of salbutamol.

In the mass spectrum of **A**, the ratio of intensities of the M^+ and $(M+1)^+$ peaks is 9.09:1, whereas the ratio of intensities of M^+ and $(M+2)^+$ is roughly 1:1.

**(i)** Determine the molecular formula of **A**. [1]

The mass spectrum of **A** also shows peaks at m/e 179 and 241.

The infra-red spectrum of **A** shows 2 peaks in the region of 1670 cm^{-1} to 1750 cm^{-1}.

(ii) Use these data, and your answer to **(c)(i)** to deduce the structure of **A**. Explain your reasoning fully. [3]

(iii) Use your structure to sketch the expected signals in the region of 3000 cm^{-1} to 4000 cm^{-1} in the infra-red spectrum of **A**. Label the peaks clearly with the corresponding functional groups and state the types of vibrations. [2]

(d) Corbadrine, a drug that is structurally similar to adrenaline, is used in the treatment of nasal congestion and vasoconstriction. Compound **L** undergoes E2 reaction when it is heated with a base to give compound **M**.
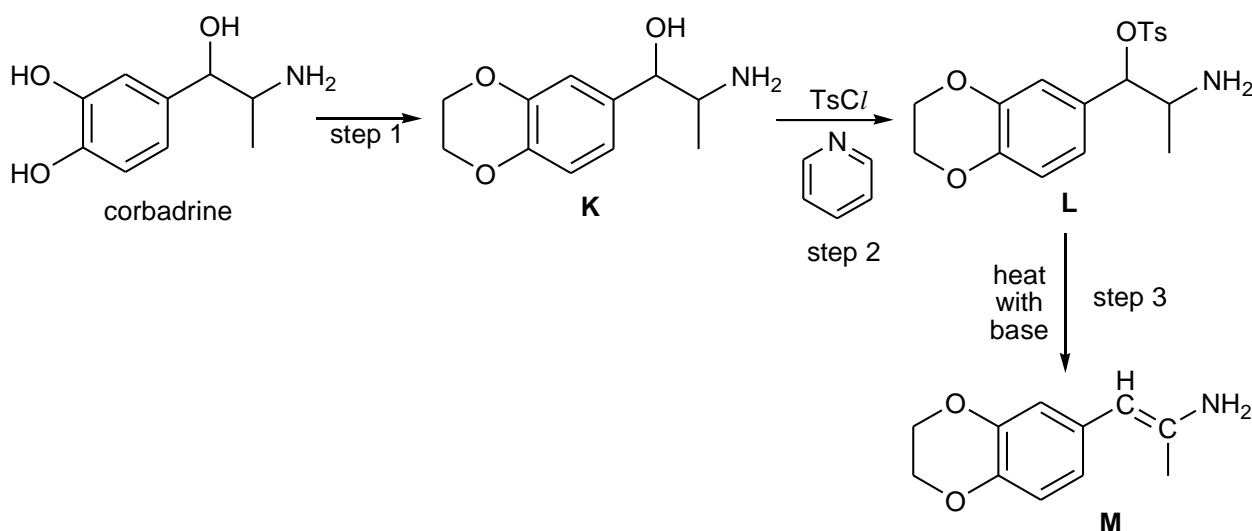
(i) State the reagents and conditions for step 1. [1]

(ii) Corbadrine exists as four optical isomers, which can be arranged in pairs of diastereoisomers. Draw the structure of **both** isomers in **one** of the pairs of diastereoisomers of compound **L**, and use the *R/S* convention to label their stereochemistries. [2]

(iii) Using the isomers you have drawn in **(e)(ii)**, draw suitable Newman projections in compound **L** to represent the conversion of **L** to **M**.

Hence predict which of your isomers give the *E* form, and which give the *Z* form, of the alkene **M**. [2]

[Total: 20]