Laboratory exercise No.3

Qualitative Analysis of Organic Compounds

A) Qualitative Analysis for Elements (Elemental analysis by mineralisation)

In organic compounds the elements commonly occurring along with carbon and hydrogen, are oxygen, nitrogen, sulphur, chlorine, bromine and iodine. The detection of these elements in organic compounds depends upon **converting them to water-soluble inorganic compounds** and the application of specific tests (the conversion is called „mineralisation"). There can be performed an oxidative mineralisation (e.g. catalysed with CuO) or reductive mineralisation (with Na or Mg).

Lassaigne's test after reductive mineralisation

C, H, O, N, S, X -> X- CN- S^{2-} CNS-

Fusion tube

PROCEDURE

Place a little of the compound (50 mg or 2 - 3 drops) into a fusion tube. Add a reaction mixture (magnesium powder + potassium carbonate) in the fusion tube tip but keep there a gap for hot air to escape.

Heat the fusion tube gently at first by flame of a gas burner. Then heat up the reaction mixture. When charring begins, heat the bottom of the tube to dull redness (but avoid melting of the glass!). Finally plunge the tube, while still hot, into a clean dish test tube half-filled with cold distilled water (cca 6 mL) kept in a test tube stand. The fusion tube breaks and the content is released into the water. Filtrate the mixture. **The 'fusion' filtrate** which should be clear and colourless, is used for the SPECIFIC TESTS DESCRIBED BELOW:

1. NITROGEN (CYANIDES)

To a portion (2 mL) of the 'fusion' filtrate add 0.2 g of powdered ferrous sulphate crystals. If also sulphide S^{2-} is present, black precipitate of FeS is formed. Boil the mixture for a half a minute, cool and acidify by adding dilute sulphuric acid dropwise. Formation of a **bluish-green precipitate** (Prussian blue) or a blue solution indicates that the original substance contains nitrogen. If no precipitate appears, allow to stand for 15 minutes, filter and inspect filter paper.

$$Fe^{3+} + Fe^{2+} + 6\ CN^- \rightarrow Fe^{III}[Fe^{II}(CN)_6]^-$$

2. SULPHUR (SULPHIDE)

To the cold 'fusion' filtrate (1 mL) add a few drops of cold, freshly prepared, dilute solution of sodium nitroprusside. The latter may be prepared by adding a small crystal of the solid to 2 mL of water. Production of a **rich purple** colour indicates that the original substance contains sulphur. This test is very sensitive.

$$S^{2-} + [Fe(CN)_5NO]^{2-} \rightarrow [Fe(CN)_5NOS]^{4-}$$

3. HALOGENS (HALIDES)

Acidify a portion (1 mL) of the 'fusion' filtrate with 2M nitric acid. Add silver nitrate (if nitrogen and/or sulphur are also present, the addition will precipitate silver cyanide and/or silver sulphide in addition to the silver halides. The removal of hydrogen cyanide and/or hydrogen sulphide is accomplished by boiling the 'fusion' solution-boil for 1 - 2 minutes).

Cool and add aqueous silver nitrate (1 mL), compare with a blank. Formation of a heavy, **white or yellow precipitate** of silver halide indicates halogen. $Ag^+ + X^- \rightarrow AgX$
If a positive result is obtained: acidify the remaining portion (1 mL) of the 'fusion' filtrate with dilute sulphuric acid, boil and cool. Add carbon tetrachloride (or chloroform) (1 mL) and a few drops of freshly prepared **chlorine water (Chloramine T solution)**. Shake the mixture.

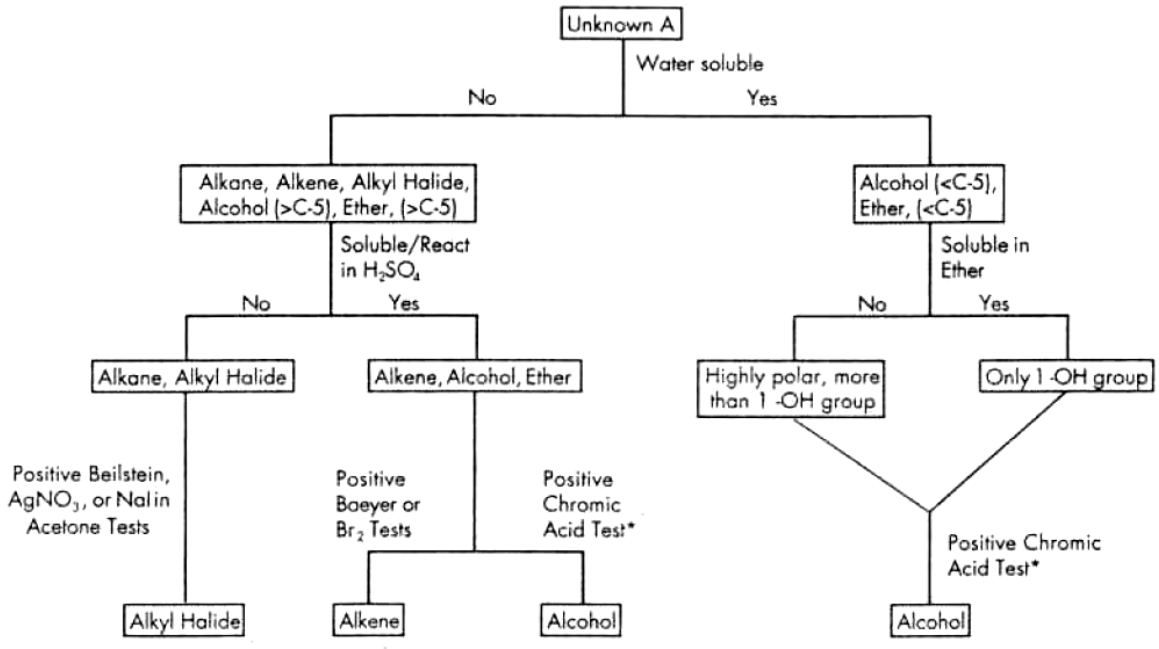
(a) If the chloroform layer remains colourless - indicates chlorine
(b) If the chloroform layer is brown - indicates bromine
(c) If the chloroform layer is violet - indicates iodine.

B) Functional analysis (Classical organic analysis of functional groups)

Elemental analysis reveals only elements (if we do quantiative elemental analysis, we can even calculate rational the formula, e.g. C_4H_5O), but nothing more. We cannot say if the compound is phenol or ether or alcohol…Therefore, we also want to know what chemical properties the analyte has, i.e. what functional groups are present. The following charts can be used for a simple classification of unknowns. The classification is based on SOLUBILITY* tests and seven classical chemical reactions.

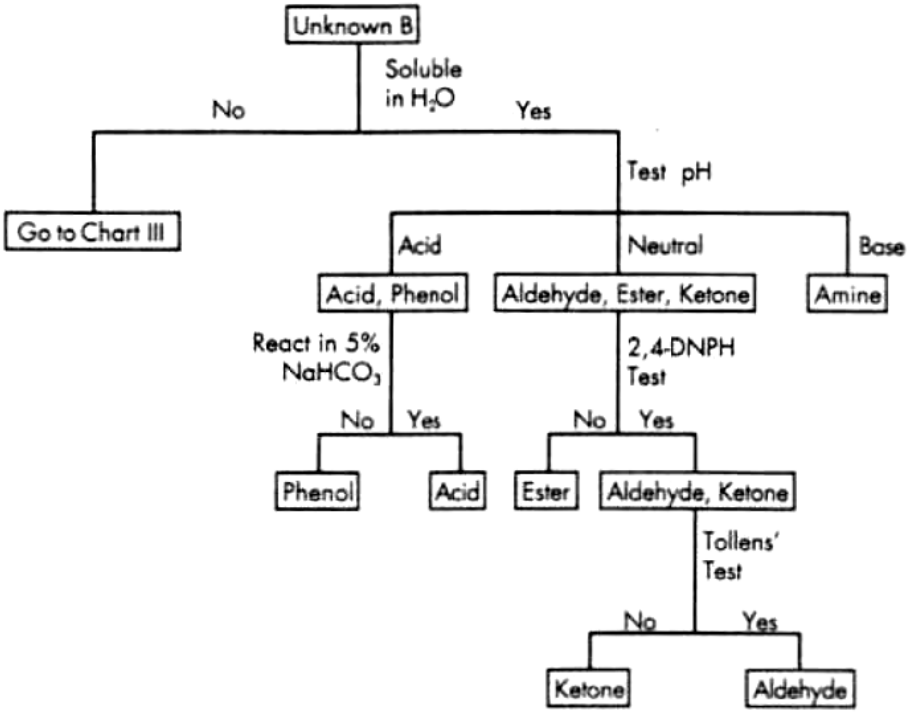
*In here, solubility is understood generally as "transformation into liquid phase", no matter if controlled by the principle *"similis similibus solventur"* or by a chemical acid-base reaction. **Solubility** is tested in a test tube: several crystals or drops of liquid is placed on the bottom of a test tube and 2-4 mL of solvent is added. Careful observation after shaking should reveal if

- crystals disappear/remain solid
- one-layer/two layers are formed



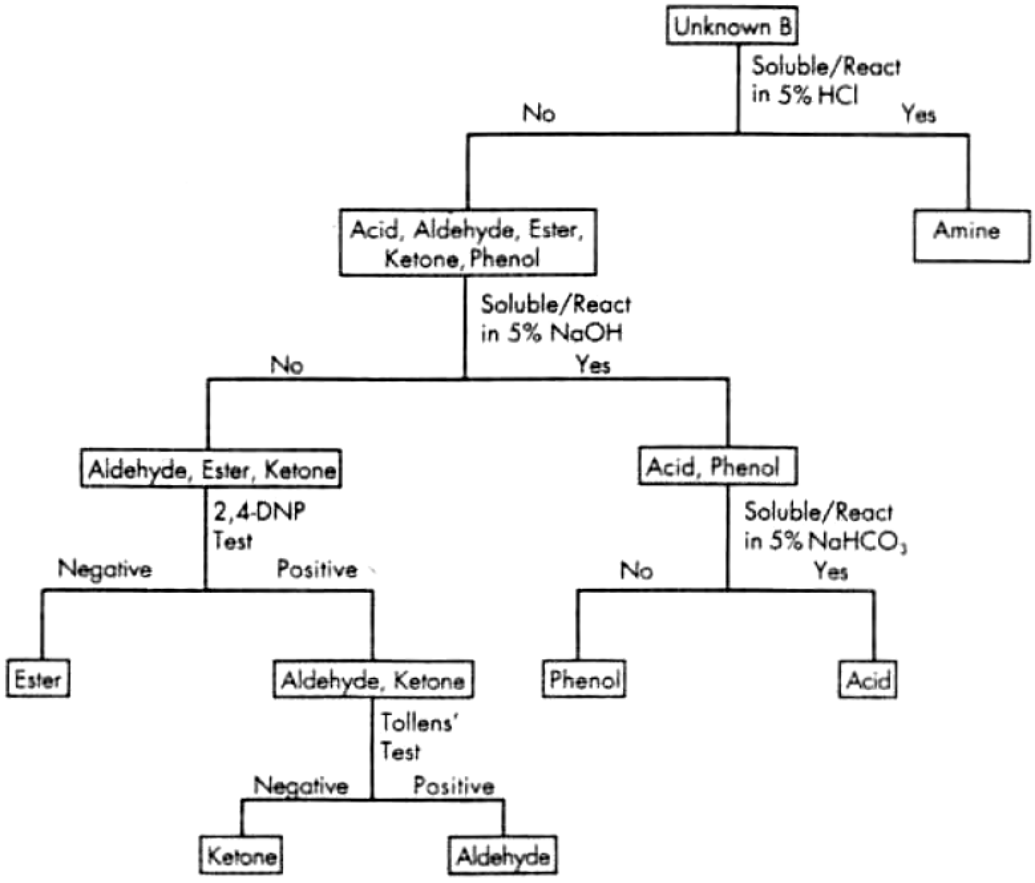
FLOW CHART
For Alkane, Alkene, Alkyl Halide, Alcohol, and Ether

Flow Chart I



Flow Chart II

For Acid, Aldehyde, Amine, Ester, Ketone, and Phenol



Flow Chart III

For Water-Insoluble Acid, Aldehyde, Amine, Ester, Ketone, and Phenol

To understand the flowcharts, we need to know of the following reactions:

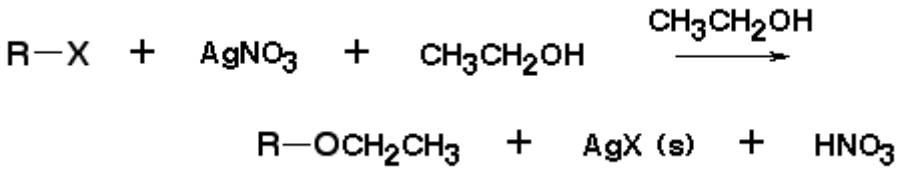
1) Beilstein test for halogenated compounds
2) Silver nitrate test in ethanol for halogens
3) Chromic acid test (Jones test) for alcohols
4) Bayer test for unsaturated compounds
5) Lucas test for alcohols classification
6) Tollens test for aldehydes
7) Reaction with 2,4-DNPH for aldehydes and ketones

1) Beilstein test for halogenated compounds

Procedure
This test is very sensitive (i.e. even impurities of halogenated compounds can give positive reaction). Take a piece of copper wire with a loop/hook on the end and heat it in the flame of a Bunsen burner until it glows red. Allow the wire to cool but avoid contaminating it (especially if you put it down on the table surface). Dip the cool loop into the known compound, and place it in the flame.
You should observe **a green flame** after the first few seconds when your known compound burns (yellow flame). Burn off all remaining halogenated known compound, heat the loop until it glows red, then let the loop cool and run the test on you unknown solid or liquid. A green flash is indicative of chlorine, bromine, and iodine, but NOT fluorine. The blue-green color is due to the emission of light from excited states of copper halide that has vaporized in the burner flame.

Standards: brom-butane, bromobenzene or chlorbenzene for the liquid and a halogenated benzoic acid for the solid.

2) Silver nitrate test in ethanol for halogenes

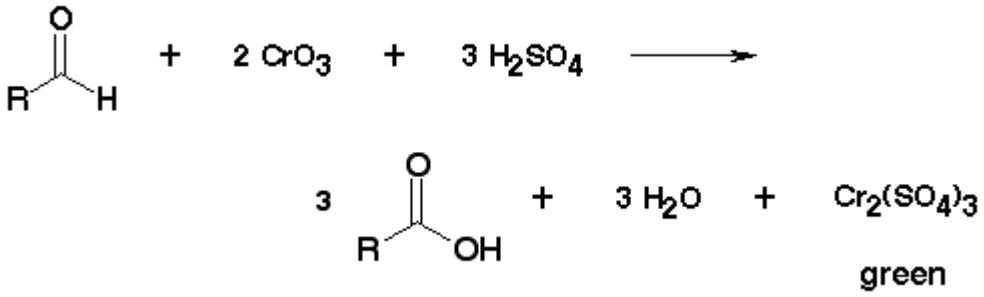


Procedure
Place approximately 0.25 mL of each compound into a test tube. Add 2 mL of a 1% ethanolic silver nitrate solution to the material in each test tube, noting the time of addition. After the addition, shake the test tube well to ensure adequate mixing of the compound and the solution. Record the time required for any precipitates to form. If no precipitates are seen **after 5 minutes**, heat the solution on the steam bath for approximately 5 minutes. Note whether a precipitate forms in the test tube.

Interferences
Carboxylic acids have been known to react in this test, giving false positive (RCOOAg).

Standards: 1-chlorobutane, 1-bromobutane, 1-iodobutane, 2-chlorobutane, 2-bromobutane, 2-iodobutane, 2-chloro-2-methylpropane, 2-bromo-2-methylpropane, benzyl chloride, bromobenzene

3) Chromic Acid Test (Jones test) for **primary** alcohols and aldehydes



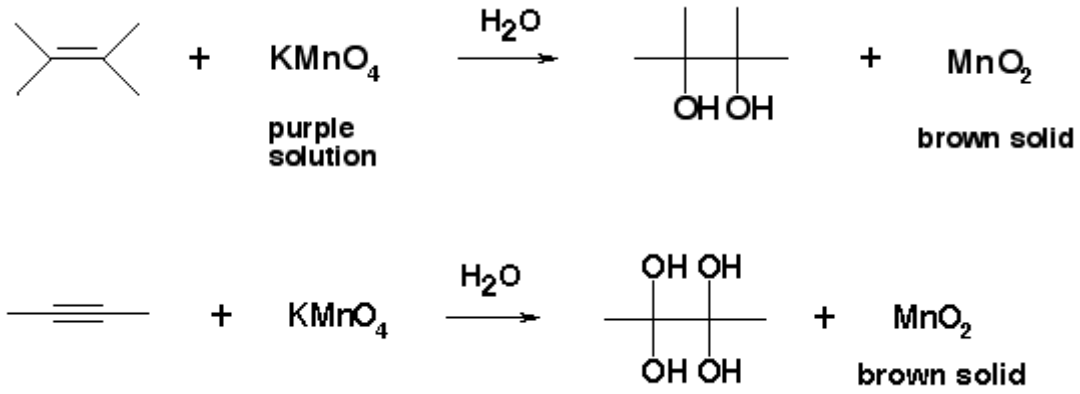
Procedure
Dissolve 10 mg or 2 drops of the unknown in 1 mL of pure **acetone** in a test tube and add to the solution 1 small drop of Jones reagent (chromic acid or $K_2Cr_2O_7$ in concentrated sulfuric acid). A positive test is marked by the formation of a green color within 5 seconds upon addition of the orange-yellow reagent to a primary or secondary alcohol. Aldehydes also give a positive test, but tertiary alcohols do not.

A positive test for aldehydes and primary or secondary alcohols consists in the production of an opaque suspension with a green to blue color. Tertiary alcohols give no visible reaction within 2 seconds, the solution remaining orange in color. Disregard any changes after 15 seconds.

Complication: the reaction is very strong oxidation (non specific). Ketones in enol form, may positively react (e.g. diketones).

Standards: Benzaldehyde

4) Bayer test for unsaturation ($KMnO_4$)



Procedure
Dissolve 1 drop or 0.02 grams of the unknown in 0.5 mL reagent grade acetone. Add a 1% aqueous solution of potassium permanganate dropwise with shaking. If more than one drop of reagent is required to give a purple color to the solution, unsaturation or an easily oxidized functional group is present. Run parallel tests on pure acetone

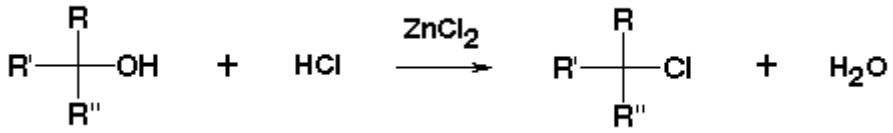
Permanganate Test for Unsaturation (Baeyer Test): Aqueous permanganate rapidly oxidizes double and triple bonds while being reduced to MnO2, a brown precipitate. Therefore, disappearance of the purple color and formation of a brown precipitate in minutes is a positive test. However, other compounds react slowly with the reagent including alcohols, aldehydes, phenols, and aromatic amines so interpret your results carefully and look for corroboration from the other tests.

Into a clean test tube, dissolve 0.1 mL of a liquid (or 50 mg of a solid) in 1 mL of 95% ethanol or 1,2-dimethyoxyethane. Add a 1% solution of aqueous potassium permanganate dropwise with agitation.

Complications: Water insoluble compounds should be dissolved in ethanol, methanol, or acetone.
Often, the brown precipitate fails to form and the solution turns reddish-brown. False positive test: easily oxidized compounds give a positive test: a) Most aldehydes give a positive test. b) Formic acid and its esters give a positive test. c) Alcohols with trace impurities give a positive test. d) Phenols and aryl amines give a positive test. False negative: carbonyl compounds which decolorize bromine/methylene chloride usually give a negative test.

Standards
Cyclohexene and Bromobenzene

5) Lucas test for tert. and sec. alcohols ($ZnCl_2$, HCl)



Procedures
To 0.2 mL or 0.2 g of the unknown in a test tube add 2 mL of the Lucas reagent at room temperature. Stopper the tube and shake vigorously, then allow the mixture to stand. Note the time required for the formation of the alkyl chloride, which appears as an insoluble layer or emulsion.

- 3^o alcohols (RRR-OH): immediate to 2-3 minutes
- 2^o alcohols (RRH-OH): 5 -10 minutes (or by heating up)
- 1^o alcohols (RH2-OH): no reaction

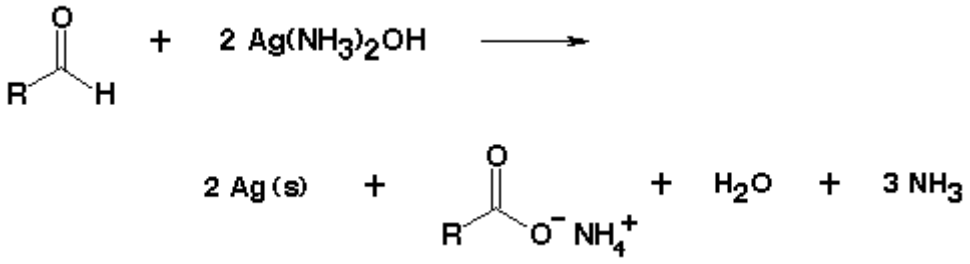
Complications: The test applies only to those alcohols soluble in water (the reagent) =monofunctional alcohols lower than hexyl and some polyfunctional alcohols. This often means that alcohols with more than six carbon atoms cannot be tested.

Standards

1-Butanol, 2-Butanol, t-Butyl alcohol

6) Tollens test for aldehydes and other easily oxidized functional groups (5%$AgNO_3$, 10%NaOH, 2% NH_3, water bath)

Tollens reagent: Into a test tube which has been cleaned with 3M sodium hydroxide, place 2 mL of 0.2 M silver nitrate solution, and add a drop of 3M sodium hydroxide. Add 2.8% ammonia solution, drop by drop, with constant shaking, until almost all of the precipitate of silver oxide dissolves. Don't use more than 3 mL of ammonia. Then dilute the entire solution to a final volume of 10 mL with water.



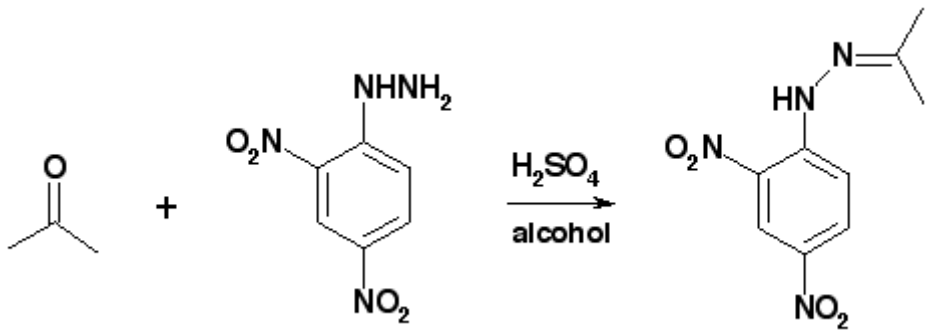
Procedure
In this test, a stabilized silver ion is reduced to elemental silver by an easily oxidized compound, such as an aldehyde. Add one drop or a few crystals of unknown to 1 mL of the freshly prepared Tollens reagent. **Gentle** heating can be employed (75°C, water bath) if no reaction is immediately observed. Formation of **silver mirror or a black precipitate** is a positive test.

Complications

- False positive: Easily oxidized compounds give a positive test. For example: aromatic amine and some phenols. Silver reduction is induced by heating **over 75°C**
- Many simple ketones give false positive reactions =Acetone, methyl ethylketone…, only isopropylmethyl ketone does not – but it does not react with 2,4-DNPH.
- Acetaldehyde does not react.

Standards: benzaldehyde

7) Reaction with 2,4 – dinitrophenylhydrazine (2,4-DNPH) for aldehydes and ketones



Aldehydes and Ketones--2,4-DNPH. Hydrazines such as 2,4-dinitrophenylhydrazine react with the carbonyl group of aldehydes and ketones to give colored precipitates. Normally, the reaction is fast but heating may be necessary. The test solution is prepared using sulfuric acid and 95% ethanol. Later, if you wish to make a derivative of your compound, you can use a different 2,4-DNPH solution prepared with HCl and methanol. This usually gives a slower forming precipitate which often provides a derivative of higher purity (and higher mp). However, the slow formation of the precipitate is not desirable when looking for a qualitative test signal. The 2,4-DNPHs are usually yellow, orange, or red with the deeper color often signifying higher conjugation via double bonds or aromatic rings. Formation of a precipitate is a positive test.

Dissolve only 1 drop of your liquid compound (or 10 mg of your solid) in a minimum number of drops of 95% ethanol in a test tube. Add 1 mL of the 2,4-DNPH *test* solution and agitate. If a precipitate does not form in 10 minutes, heat on a water or steam bath for a few minutes.

Complications
a trace of acetone will give a positive test. Some ketones give oils which will not solidify. Some allylic alcohols are oxidized by the reagent to aldehydes and give a positive test. Some alcohols, if not purified, may contain aldehyde or ketone impurities.

Standards
Cyclohexanone, Benzophenone, Benzaldehyde

END OF ANALYSIS

Progress of operations

I. Functional analysis reactions with standards (always with a blank reaction)

II. Elemental analysis of the sample A
III. Functional analysis of the sample B

ORGANIC ANALYSIS DATA TABLE

	Known			Unknown	
	Procedure compound	**Results**	**Conclusion**	**Results**	**Conclusion**
1	Beilstein				
2	$AgNO_3$				
3	Jones *ethanol*				
4	Bayer				
5	Lucas				
6	Tolens				
7	2,4-DNPH				