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A REVIEW: REAGENT IN PHARMACEUTICAL ANALYSIS

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ABSTRACT

In pharmaceutical analysis both for qualitative and quantitative analysis reagents are important, various reagents are routinely used in qualitative and quantitative analysis of pharmaceuticals. In this section we focussed on principles and procedures of some important reagents used in pharmaceutical analysis. 1, 2-naphthoquinone-4-sulfonate is a chemical regent used to measure levels of amines and amino acids like ethylenimine limit test, colorimetric determination of cefadroxil and ceftriazone in pharmaceutical dosage forms and spectrophotometric determination of pregabalin using 1, 2-napthaquinone-4-sulfonic acid sodium. 2,3,5-triphenyltetrazolium is redox indicator used in Seed analysis and Quantitative determination of effects of Mycoplasma pneumonia cells and Membranes.(study of the infective process in primary atypical pneumonia) and so many regent used in pharmaceutical analysis. 3-methylbenzthiazolinone-2(3H)-hydrazone originally introduced as a reagent for aldehyde Analysis of Acyclovir, Analysis of Ceftazidime, Analysis of Ganciclovir and Analysis of Cefadroxil.

Keywords: 1, 2-naphthoquinone-4-sulfonate, 2, 3, 5-triphenyltetrazolium, 3-methylbenzthiazolinone-2(3H)-hydrazone, Cefadroxil, Mycoplasma pneumonia and Ceftazidime.

INTRODUCTION

A reagent is a "substance or compound that is added to a system in order to bring about a chemical reaction, or added to see if a reaction occurs".

Although the terms reactant and reagent are often used interchangeably, a reactant is more specifically a "substance that is consumed in the course of a chemical reaction". Solvents, although they are involved in the reaction, are usually not referred to as reactants. Similarly, catalysts are not consumed by the reaction, so are not described as reactants.

In organic chemistry, reagents are compounds or mixtures, usually composed of inorganic or small organic molecules that are used to effect a transformation on an organic substrate. Examples of organic reagents include the Collins reagent, Fenton's reagent, and Grignard reagent. There are also analytical reagents which are used to confirm the presence of another substance. Examples of these are Fehling's reagent, Millon's reagent and Tollens' reagent.

In another use of the term, when purchasing or preparing chemicals, reagent-grade describes chemical substances of sufficient purity for use in chemical analysis, chemical reactions or physical testing. Purity standards for reagents are set by organizations such as ASTM International or the American Chemical Society. For instance, reagent-quality water must have very low levels of impurities like sodium and chloride ions, silica, and bacteria, as well as a very high electrical resistivity.

1,2-NAPHTHOQUINONE-4-SULFONATE

Folin's reagent or sodium 1,2-naphthoquinone-4-sulfonate is a chemical reagent used to measure levels of amines and amino acids shows in figure 1. The reagent produces a bright red colour in alkaline solutions and is also fluorescent [1].

This should not be confused with Folin-Ciocalteu reagent, which is a mixture of sodium tungstate and sodium molybdate that is used to detect phenolic compounds [2].

Ethylenimine Limit Test

Scope

This procedure is designed to detect the presence of ethylenimine in immobilized enzyme preparations containing poly (ethylenimine).

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Principle

The principle of the method is to react any free ethylenimine which may be present in a sample of immobilized enzyme preparation with an aqueous solution of 1,2-naphthoquinone- 4-sulfonate (Folin's reagent) to produce 4-(1-aziridinyl)-1,2-naphthoquinone. This reaction product is extracted into chloroform and the extract analyzed by high performance liquid chromatography (HPLC) [3].

Reagents and Solutions

- Chloroform with 1% ethanol as a stabilizer,
- Hexane,
- 2-propanol,
- Methyl alcohol
- Acetone,
- 1,2-naphthoquinone-4-sulfonic acid, sodium salt
- N sodium hydroxide (NaOH)
- M Potassium dihydrogen phosphate (KH2PO4)
- 4-(1-Aziridinyl)-1,2-naphthoquinone

Buffer Solution: pH 7.7; mix 200 ml of 0.1 M KH2PO4 with 93.4 ml of 0.1 N NaOH.

Folin's Reagent: Dissolve 0.40 g of 1,2-naphtoquinone-4-sulfonic acid sodium salt in 100 ml of buffer solution. Dilute to 500 ml with distilled water in a volumetric flask.

4-(1-Aziridinyl)-1,2-naphthoquinone

A standard sample of known purity is required. If a commercial source for this standard is not readily available, the substance may by synthesized by the following procedure:

- Wrap a separatory funnel with aluminium foil and add 2 g of the sodium salt of 1,2- naphthoquinone-4-sulfonic acid dissolved in 250 ml of distilled water.
- Add 25 ml of 0.5 M trisodium phosphate shake and check that the pH is between 10.5 and11.5. Add 0.3 ml ethylenimine and shake intermittently for 10 min.
- Extract the 4-(aziridinyl)-1,2-naphthoquinone formed with six 200-ml portions of chloroform.
- Place the combined extracts in a 2-liter beaker wrapped in aluminium foil in which three holes have been made.
- Evaporate the chloroform at room temperature with a nitrogen purge. Transfer the dry residue to a 50-ml beaker wrapped in aluminium foil.
- Add 35 ml of methyl alcohol and 1 ml of chloroform to the residue and stir briefly. Not all of the residue will dissolve.
- Place the beaker in an ice-water bath for 10 min and then filter the precipitate through Whatman 42 filter paper.
- Rinse the precipitate in the filter with 4 ml of chilled methyl alcohol and discard the filtrates.
- Dry the precipitate with a nitrogen purge, transfer it to a brown glass bottle and purge again.
- Dry the compound overnight in a desiccator containing Drierite. The melting point of the compound is 173-175°. The compound is to be used for making standard solutions for calibration purposes. The compound should be stored in a freezer until standard solutions are to be prepared.

Standard Solution

0.5 g/l Standard Solution: Accurately weigh about 125 mg of 4-(1-aziridinyl)-1,2- naphthoquinone into a 250 ml volumetric flask [low actinic glass] and add chloroform to the mark.

0.1 mg/l Standard Solution: By appropriate dilution(s) of the 0.5 g/l Standard Solution, prepare a standard solution which contains 0.1 mg/L (0.1 ng/µl).

Analysis

Accurately weigh a sample of immobilized enzyme preparation containing about 10 g of dry matter into an aluminium foil-covered beaker. Add 50 ml of Folin's Reagent and agitate the mixture for several minutes. Decant the Folin's Reagent into a separatory funnel and extract with 2 ml of chloroform. Analyze a 20 µl portion of the chloroform extract by the following chromatographic conditions:

Column: Lichrosorb DIOL 5 nm (or equivalent)

Mobile phase: hexane:chloroform (with 1% ethanol) : isopropanol = 59.5 : 40.0 : 0.5 (v/v)

Flow rate: 2 ml/min.

Inject a 20 µl portion of the 0.1 mg/L Standard Solution. The sample response is not greater than that of the 0.1 mg/L Standard Solution. Amount of ethylenimine in immobilized enzyme sample determine by calibration curve.

Colorimetric Determination Of Cefadroxil And Ceftriazone In Pharmaceutical Dosage Forms

Principle

The method was based on the derivatization of cefadroxil and ceftriaxone with 1, 2-naphthaquinone-4-sulfonic acid sodium in alkaline medium to yield orange-colored products. 1, 2-naphthoquinone-4-sulphonate (NQS) is used as a chromogenic reagent in the development of a simple and rapid colorimetric method for the determination of CFL and CFX in pharmaceutical dosage forms. The absorbance of the reaction solution increased as NQS concentration increased, and the highest absorption intensity was attained at NQS concentration of 0.25 % w/v. To generate nucleophiles from CFL/CFX and activate nucleophilic substitution reactions, alkaline medium is necessary. So 0.01M sodium hydroxide is also used [4,5].

Proposed reaction mechanism for the formation of coloured species show in figure 2,3.

Procedure

Reagents and solutions

- 1, 2-Naphthoquinone-4-sulphonate (NQS) 0.5 %w/v
- Sodium hydroxide solution (0.01 M)
- Standard solutions of CFL and CFX

Preparation of calibration curve

Standard solutions of CFL and CFX in water, having final concentrations in the range of 10 - 100 µg/ml and 25 - 175 µg/ml, respectively, were transferred into a series of 10 ml volumetric flasks. To each of these solutions, 1 ml of 0.01M sodium hydroxide was added, followed by 1 ml of 0.5 % NQS. The mixture was then gently shaken until the appearance of orange colour. The contents were diluted to 10 ml with distilled water. The absorbance of each solution was measured at 475 nm and 480nm for CFL and CFX, respectively, against the reagent blank prepared in the same manner, without the analyte and calibration curve is plotted [6-7].

Determination of CFL and CFX

Sample solution of CFL and CFX of 50 µg/ml and 150 µg/ml concentration respectively are taken. To the sample Solutions of CFL(0.5ml) and CFX(1.5 ml) , 1 ml of 0.01M sodium hydroxide was added, followed by 1 ml of 0.5 % NQS. This both mixture were then gently shaken until the appearance of orange colour. The contents were diluted to 10 ml with distilled water and the absorbance of each solution was measured at 475 nm and 480nm for CFL and CFX, respectively, against the reagent blank prepared in the same manner, without the analyte. The concentration of CFL and CFX in each standard flask was obtained by interpolating the corresponding absorbance value of the product from Beer's plot of standard CFL and CFX solutions.

Spectrophotometric Determination of Pregabalin Using 1, 2-Napthaquinone-4-Sulfonic Acid Sodium

Pregabalin (PGB) [S-[+]-3-isobutyl GABA or (S)-3-(amino methyl)-5-methylhexanoic acid, Lyrica] is an anticonvulsant and analgesic medication that is both structurally and pharmacologically related to gabapentin. NQS also used as chromogenic reagents in the development of simple and rapid spectrophotometric method for the determination PGB in its pharmaceutical dosage forms [8-10].

Principle

The NQS reagent reacts with PGB at the free NH2 group represented in structure .NQS reagent forms colored complexes with PGB in alkaline conditions and their absorbance was measured at 485nm reaction show in figure 4. Because of the presence of amine as chromophoric group in the PGB molecule, derivatization of the compound was attempted with NQS as a result colored complex has been formed which was estimated spectrophotometrically

Procedure

Preparation of calibration curve

Standard solutions of PGB in water, having final concentrations in the range of 5-50 µg/ml was transferred into a series of 10 ml volumetric flasks. To these solutions, 1 ml of 0.01N sodium hydroxide is added, 1 ml of 0.5% NQS is added. The mixture was then gently shaken until the appearance of orange colour. The contents were diluted up to 10 ml with distilled water. The absorbance of each solution was measured at 485 nm against the reagent blank and the calibration curve is plotted [11, 12].

Analysis

Sample solution of PGB of 40 µg/ml concentration is taken. To the sample Solutions of PGB , 1 ml of 0.01N sodium hydroxide was added, followed by 1 ml of 0.5 % NQS. the mixture is then gently shaken until the appearance of orange colour. The contents were diluted to 10 ml with distilled water. And The absorbance of solution was measured at 485 nm against the reagent blank prepared in the same manner, without the PGB. The concentration of PGB is obtained by interpolating the corresponding absorbance value of the product from Beer's plot of standard PGB solutions.

2,3,5-TRIPHENYLTETRAZOLIUM

Triphenyl tetrazolium chloride, TTC, or simply Tetrazolium chloride is a redox indicator commonly used in biochemical experiments especially to indicate cellular respiration show in figure 5. It is a white crystalline powder, soluble in water, ethanol and acetone.

Tetrazolium salts are colorless indicators that are reduced to form intensely colored, relatively insoluble compounds, i.e. formazans by accepting electrons from suitable donors, normally metabolically active cells which contain dehydrogenases enzyme such as numerous types of cells, including leukocytes, fungi, bacteria, neurons, and Ehrlich ascites tumor cells. The enzymes responsible for the reduction are variety of dehydrogenases and thus activity is associated primarily with viable (active) cells.

Principle

TTC (2,3,5-triphenyltetrazolium chloride) is a redox indicator used to differentiate between metabolically active and inactive tissues. The white compound is enzymatically reduced to red TPF (1,3,5 triphenylformazan) in living tissues due to the activity of various dehydrogenases (enzymes important in oxidation of organic compounds and thus cellular metabolism), while it remains as white TTC in areas of necrosis since these enzymes have been either denatured or degraded [13-15].

ANALYSIS USING TTC REAGENT

Seed Analysis (TZ Test) For Seed Viability Principle

The TZ test estimates seed viability. Prepared seeds are soaked in a solution of water with 2,3,5 triphenyl tetrazolium chloride. The abbreviation TTC is used for the chemical while the abbreviation TZ is used for the solution and the test in general. In the living tissues of the seeds, active enzymes (dehydrogenases) that produce free H+ ions convert the TTC chemical to formazan, an insoluble red dye that stains the living tissues red show in figure 6. The reaction takes from a few hours to one or two days, depending on the type of seed and other factors.

TZ results are expressed as percent viability. The TZ test becomes a dormancy test when it is done on the ungerminated seeds left after a standard germination test or as a separate test alongside the germination test. A TZ test alone cannot measure dormancy. TZ tests can also provide vigor information when the reaction variables are tightly controlled and the analyst uses check samples (samples of known high vigor). The nature and extent of mechanical damage, thermal damage, aging, embryo maturity and insect and fungal pathogens can also be detected with a TZ test.

Procedure

There are five steps in a TZ test. Some steps may be omitted depending on the species of the seed.

1. Preconditioning: Most seeds need to be imbibed with water before they are cut or prepared for the staining solution. Seeds are placed on moistened germination media (blotters or towels) overnight or for several hours.
2. Preparation: The imbibed seeds are then pierced or cut to expose the embryo to the staining solution. Some seeds require removal of covering structures like the fruit coat or the seed coat. Other species can simply be placed directly into the stain without any preparation.
3. Staining: The prepared seeds are placed into a TZ solution that has an appropriate TZ concentration for the species being tested and the preparation method chosen. Generally this is a one percent solution for whole or pierced seeds and a one-tenth percent solution for seeds that have been cut to expose the embryo. The time and temperature for staining are other variables that are species specific.
4. Preparation for evaluation: Again, this is species specific. Some dissection may be needed after staining to more clearly see the embryo structures. Lactic acid or glycerol may be used to "clear" seeds with dark seed coats if dissection is too difficult or time consuming. These clearing solutions lessen the chance of artifacts from the preparation method (2).
5. Evaluation: The analyst must know the seed structures and be able to evaluate staining patterns of the essential structures. Some parts of the seed naturally do not stain. For example, seeds in the grass family have a non-staining endosperm. The stain color itself also has to be evaluated. A bluish purple color may indicate frost damage, while an orange red may indicate that the stain is improperly buffered. Staining may be uneven, due to the rate of solution uptake or the uneven metabolic activity in the different parts of the seed. The soundness and turgidity of the tissues are also examined.

Quantitative determination of effects of mycoplasma pneumonia cells and membranes (study of the infective process in primary atypical pneumonia)

Principle

Mycoplasma pneumoniae, currently the only proven mycoplasma pathogen for humans, has a predilection for attachment to ciliated respiratory epithelium. This, plus the fact that M. pneumoniae causes pneumonia in hamsters permitted the development of the hamster trachea organ culture as a model system for the study of the infective process in primary atypical pneumonia [16-17].

The ability of Mycoplasma pneumoniae cells and membranes to affect tetrazolium reduction by hamster trachea organ cultures was evaluated. Uninfected trachea explants reduced 2,3,5-triphenyl tetrazolium chloride (TTC). Reduced tetrazolium salts (formazans) were extractable with acetone or ethylene glycol and could be quantitated spectrophotometrically by determining optical density at 490nm Tracheas exposed to mycoplasma cells or membranes showed significantly decreased ciliary activity and tetrazolium reduction; e.g., only 5% of the ciliary activity and reduction capacity remained after 5 days in culture when infected with M. pneumonia PI 1428 cells [18].

The data indicate that the exposure of ciliated respiratory epithelium to mycoplasma cells or membranes results in diminished oxidative metabolism, and that the ability to reduce TTC to its formazan is correlated with relative ciliary activity.

Procedure

- M. pneumoniae, strains FH was taken.
- Solutions of 0.12% 2,3,5-triphenyl tetrazolium chloride (TTC) were made in Tyrode balanced salt solution supplemented with 1.2% sodium succinate.
- Trachea rings were incubated for 2 h in 1.0 ml of the tetrazolium mixture at 37 C in the dark.
- Low oxygen conditions were obtained by incubating the vials (16 by 60 mm) in a desiccator evacuated and flushed (two cycles) with 100% nitrogen, or by overlaying the solutions with 1.0 ml of mineral oil.
- After the incubation period, fluids were aspirated and the rings were blotted dry.
- Formazan was extracted with 1.0 ml of acetone for 5 min.
- The optical density at 490 nm (OD_{490}) of the acetone solution was measured spectrophotometrically.
- Dry weights of the tracheas were determined, and the OD_{490} per milligram was calculated.

RESULTS

The data show a rapid drop in relative activity for the infected rings. By day 5, activity had dropped to approximately 5% of the control level (relative activity = 254). On day 5, tetrazolium reduction was also measured, and significant differences were found between control and infected rings. The OD_{490} per milligram (dry weight) readings for controls were 1.215, compared with a value of 0.272 for the mycoplasma-infected explants thus total 78% decrease.

3-METHYL-2-BENZOTHIAZOLINONE HYDRAZONE (MBTH REAGENT)

3-methylbenzthiazolinone-2(3H)-hydrazone originally introduced as a reagent for aldehyde. Later its use was extended to a variety of organic compound (example: Phenols, aryl amines and different N- and S-heterocyclic compound) figure 7. The reagent is used in the form of an aqueous solution and the reaction products can be extracted into chloroform, if desired [19-20].

Applications

1. MBTH reagent is used in the analysis of drinking surface and saline waters, domestic and industrial wastes.
2. MBTH reagent is also used in measuring phenolic material at the 2 mcg/L level, when the colored end product is extracted and concentrated in a solvent phase using phenol as a standard.
3. Used for the determination of drugs like acyclovir, ganciclovir, ceftazidime and cefradoxil, nicorandil.
4. Used in the identification of groups such as
- Aldehydes
- Amines
- Phenols
- Aryl Amines

5. The MBTH procedure was also adapted for use in seawater samples to measure the formaldehyde given off by the marine methanotroph, Methylomonas pelagic
6. MBTH used for estimation of samples which contain higher concentrations of aldehydes such as disinfectants.

Principle of MBTH reagent

MBTH reacts with aldehyde first to form an azine. Only if there is remaining MBTH, it is oxidized to another species which combines with the azine to form formazan. However, if there is enough aldehyde, all the MBTH is converted to azine and there is no formation of blue color. Thus, by using the limiting agent MBTH to test the amount of aldehyde around the point of interest, then less aldehyde would produce more blue color reaction show in figure 8 and more aldehyde would produce less blue color. The end color may be different depending upon the order of addition of the reactants. For example, if an oxidizing agent and MBTH are mixed before adding the aldehyde, a light green to green/blue color results. This method could be used for solution test or for tests on a solid phase such as on nylon membrane. The latter also could be used for measurements with a device or instrument such as a color reader and used in combination with a second aldehyde tester and a pH tester.

With Phenols under reaction condition MBTH loses two electrons and one proton to form the electrophilic intermediate, which has been identified as the active coupling species that undergoes electrophilic substitution with phenol and other groups to form the colored product.

ANALYSIS OF VARIOUS DRUGS USING MBTH REAGENT

Analysis of Acyclovir Analysis

- MBTH (1 ml) was transferred into a series of 10-ml volumetric flasks.
- Aliquot volumes of acyclovir standard solution was added so that the final concentration was in the range of 20-200 mg/ml, then 1 ml of ferric chloride solution was added.
- This solution was mixed and allowed to stand for 20 min. The volume was adjusted to the mark with water.
- The absorbance was measured against a reagent blank (which contains all reagents except acyclovir) at 616 nm.
- The absorbance versus the final concentration was plotted to get the calibration curve, or to derive the regression equation.

Procedure for the test tablets

Ten tablets were weighed and pulverized. A weighed portion of the powder equivalent to 25 mg of acyclovir was transferred into 50-ml standard flask and shaken using a sonicator for 15 min. The volume was adjusted to the mark with water, mixed and filtered and then proceeded as above. The nominal content of the tablets was determined either from the calibration curve or using the regression equation.

ANALYSIS OF CEFTAZIDIME

Standard solutions

- A stock solution of ceftazidime (1 mg/mL) in water was kept in dark to avoid drug degradation. The working standard solution of ceftazidime containing 100 mg/mL was prepared by dilution.
- Aqueous solutions (0.2%) of MBTH were pepared.

Analytical procedures

- Aliquots of the working standard solution of the drug (0.2–1.0 mL) (100 mg ml-1) were transferred into 10-mL calibrated flasks.
- To each aqueous solution of FeCl3(1.5 mL, 3*10-2 mol L-1), an aqueous solution of MBTH (1.0 mL, 8.6 * 10-3 mol L-1) was added. The solutions were swirled and allowed to stand for 5 min.
- Then HCL (1.0 mL, 1*10-2 mol L-1) was added and

made up to the mark with water.

- The absorbance was measured at 628 nm against the corresponding reagent blank and calibration graph was constructed.

ANALYSIS OF GANCICLOVIR

Standard solution

Accurately weighed 100 mg of Ganciclovir was dissolved in 100 ml of distilled water and further diluted with sufficient quantity of distilled water (i.e.1000 µg/mL).

Preparation of Sample

For the estimation of Ganciclovir 20 capsule were weighed and triturate to fine powder. Capsule powder equivalent to 100mg of Ganciclovir was weighed, dissolved in 40 ml of distilled water and further diluted with sufficient quantity of distilled ethanol. This was then filtered through whatman filter paper no. 41 to get the stock solution of concentration 100 µg/mL. Further dilution was made with distilled ethanol to get the concentration of 100µg/mL.

Method

- Fresh aliquots of Ganciclovir ranging from 0.5 to 2.5 mL (1 µg/mL-1000µg/mL) were transferred into a series of 10 mL volumetric flasks to provide final concentration range of 50 to 250µg/mL.
- To each flask 1ml of aqueous Ferric chloride (1%) solution and 1 ml of MBTH reagent (0.5% in distilled water) were added. The solution in each tube were made upto the mark with distilled water.
- The absorbance of bluish green colour chromogen was measured at 611.6 nm against the blank.
- The amount of Ganciclovir present in the sample solution was computed from its calibration curve.

ANALYSIS OF CEFADROXIL

Preparation of Standard

An accurately weighed amount of CFL was dissolved in water and diluted stepwise to obtain working standard solutions of concentration 100µg/ml.

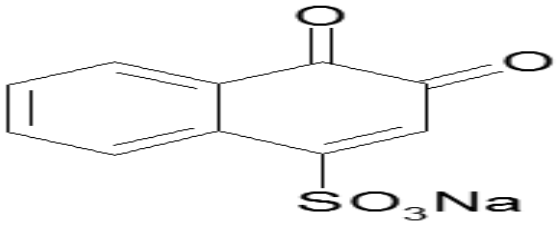
Preparation of Sample

An accurately weighed quantity of tablet or capsule powder equivalent to 50 mg of active ingredient was extracted with distilled water (2 x 20 ml) and filtered. The volume of the combined extract was brought to 50 ml with water. The filtrate (1 mg/ml) was diluted stepwise with distilled water to give the working ranges 100 µg /ml.

Procedure

To a series of 25-ml calibrated flasks, containing aliquots of CFL (0.5-3.0 ml, 50 µg/ml), 2.0 ml portions of MBTH solution were added and kept for 2 min at room temperature. Then 2.0 ml of Ce(IV) solution was added, kept for 15 min and diluted to the mark with water. The absorbencies were measured within 45 min at 410 nm against a reagent blank. The CFL concentration was read from a calibration graph prepared under identical conditions.

Fig 1. Sodium 3,4-Dioxo-3,4-Dihydrona Phthalene -1-Sulfonate



Analysis using 1,2-Naphthoquinone-4-Sulfonate Reagent

Fig 2. Cefadroxil- NQR coloured complex

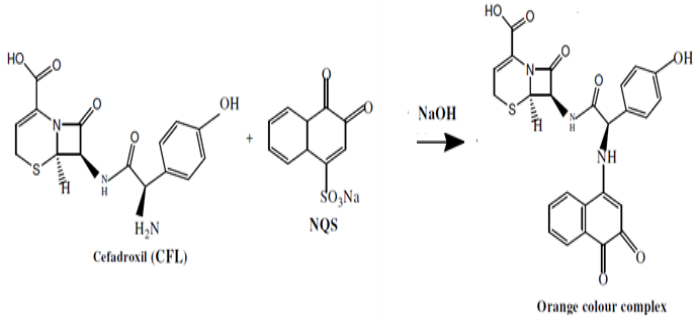


Fig 3. Ceftriaxone-NQS coloured complex

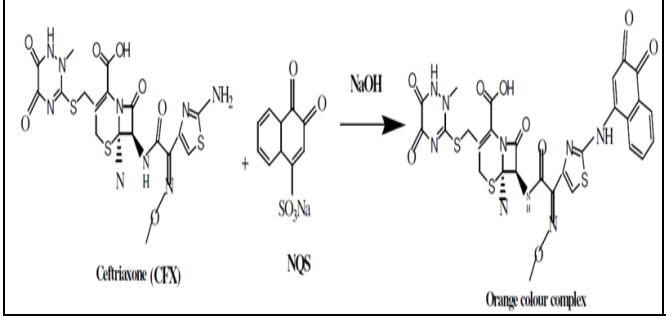


Fig 4. Pregabalin-NQS coloured complex

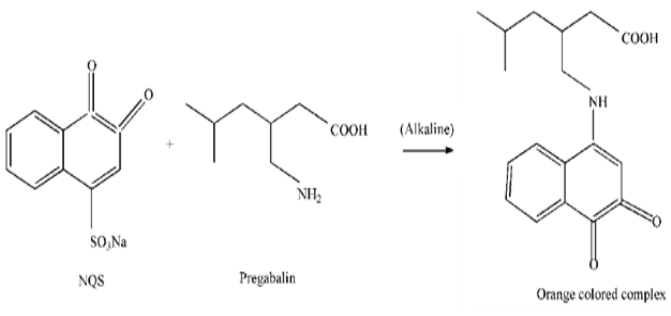


Fig 5. 2,3,5-Triphenyl-2H-tetrazolium chloride

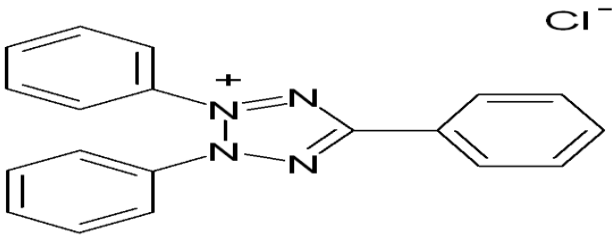


Fig 6. Colour change reaction of TPH to TPF

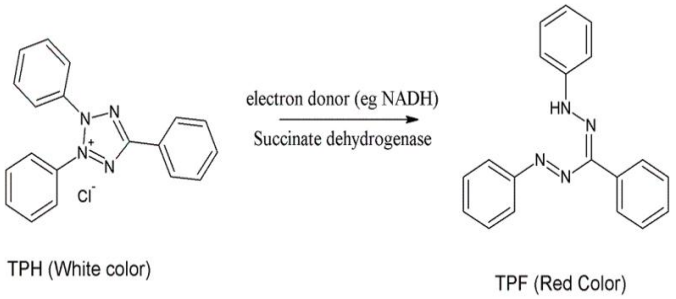
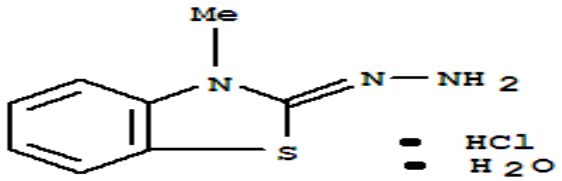
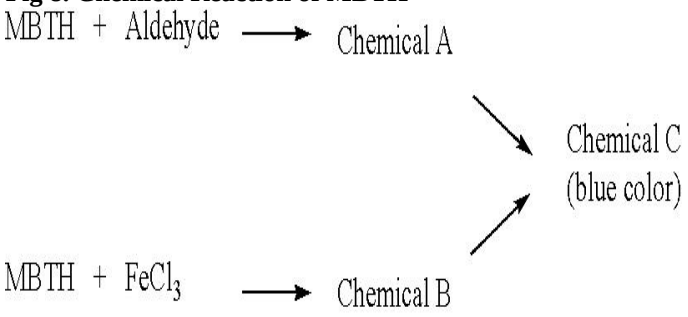


Fig 7. IUPAC Name: 3-methyl-2-benzothiazolinone hydrazine



Chemical formula: $C_8H_9N_3S.HCl$

Fig 8. Chemical Reaction of MBTH



CONCLUSION

Pharmaceutical regent are play very important role in analysis, many type of regent available in this area for use in laboratory. In this paper we discuss about some regent which is help in determination and qualitative and quantitative estimation drugs in pharmaceutical dosage from as we describe above. Folin's reagent is a chemical reagent used to measure levels of amines and amino acids and Colorimetric determination of some Cefa class of drugs and also spectrophotometric determination of Pregabalin. Tetrazolium chloride is a redox indicator commonly used in biochemical experiments in Quantitative determination of effects of mycoplasma pneumonia cells and membranes. MBTH reagent is used the determination of drugs some anti-viral drugs and cefradoxil, nicorandil.

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