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Comparative Study On Reagents Used For Extraction Of Fabric Dyes & Its Toxicity Analysis

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Abbreviations

1. UV – Ultraviolet
2. CO_2 – Carbon Dioxide
3. pH – Potential [Hydrogen]
4. EU – Europe United
5. US – United States
6. SDBS – Sodium Dodecyl benzenesulfonate
7. APEOs – Alkylphenol Ethoxylates
8. AE – Alcohol Ethoxylates
9. PEG – Polyethylene Glycol
10. APG – Alkyl Polyglycolides
11. DMSO – Dimethyl Sulfoxide
12. DEG – Diethylene Glycol
13. NMMO – N-methyl morpholine N-oxide
14. CNS – Central Nervous System
15. DNA – Deoxyribonucleic Acid
16. RNA – Ribonucleic Acid
17. NaOH – Sodium Hydroxide
18. HCL – Hydrochloric Acid
19. $NaHSO_3$ – Sodium Hydrogen Sulphite
20. Na_2CO_3 – Sodium Carbonate
21. $Na_2S_2O_5$ – Sodium Metabisulfite
22. H_2O_2 – Hydrogen Peroxide
23. Na_2CO_3. $1.5H_2O_2$ – Sodium Percarbonate
24. $NaBO_3$ $.4H_2O$ – Sodium Perborate
25. $Na_2S_2O_4$ – Sodium Dithionite
26. NH_4OH – Ammonium Hydroxide
27. N_2S – Sodium Sulphide
28. CH_3NaO_3S – Sodium Formaldehyde Sulfoxylate
29. Na_2SO_4 – Sodium Sulphate
30. $C_{14}H_{10}O_4$ – Benzoyl Peroxide
31. C_3H_6O – Acetone

32. TLC – Thin Layer Chromatography
33. PPE – Personal Protective Equipment

ABSTRACT:

Dyes have been an intriguing subject of matter and debate since its invention. Surely, it has many advantages and its usage is wide in the fields of chemistry, biology, & day-to-day life. However, there are yet; many disadvantages and the most dangerous one is its toxic nature. Modern synthetic dyes are made from chemicals which include many toxins that might harm human life and the surrounding environment. The extensive use of dyes in fabric products is also an upsurge and it's a developing problem. Yet, not many researches have been performed to target its authenticity and safety.

Even when forensic scientists compare dyes and create report or analyse individual components of dyes, it is important to extract it out safely from easy methods. My research deals with finding fast, efficient and easy method to extract fabric dyes from clothes. Fabric dyes are of many types and each different type has its own method of extraction. I have used Hot Alkaline Extraction Technique to extract dyes from cotton and polyester fabrics through different reagents and solvents and found the best reagent and solvent combination to extract the dye efficiently. My comparative analysis gives us suitable methods and best method for specific variety of clothes and helps in giving fast as well as efficient extraction tool for fabric dye extraction.

My basic research question was to find the best suitable and efficient tool for extraction of fabric dyes and determine the best reagent-solvent combination for the same. The secondary objective was to perform heavy metal and toxin test on the dye obtained to qualify the substances which might be harmful and present in the dye. Subsequently, TLC has also been done to get results of suitable mobile phase and Rh Factor.

CHAPTER 1: INTRODUCTION

1.1 General Introduction to Dyes

Dyes are substances that impart color. However, these substances may contain toxic compounds which can cause mild or severe problems to the human body. These dyes are exploited in the market to impart color in different substances including food, toys, plastic, cosmetics and even clothes. Dyes are usually classified into two types on the basis of its occurrence:

- **Natural Dyes** – Obtained and extracted from natural sources.
- **Synthetic Dyes** – Chemically formulated for cheap and extensive use.

Furthermore, Dyes can also be classified on the basis of its usage. Those dyes which are not harmful to the human body are safe to use in food, cosmetics and clothes. Those which are toxic to the body can only be used in products which are not meant for human use; e.g. plastic, containers, toys, etc. When we talk about different dyes in cosmetic as well as fabric use or edible use; there are some dyes which are expensive due to its property, extraction or any other reason. Manufacturers avoid using such expensive dyes and use cheap products which impart color easily as additives. The cheap products become a home of toxins and can impart heavy metals as well as toxic substances on humans.

Hair dyes are a hot favorite topic when toxins released from dyes are talked about. Many cheap quality hair dyes contain lead and other heavy metals which cause tremendous damage and irritation on scalp and human body. There are also edible dyes which contain harmful additives, which in order create trouble with the GI Tact and other internal organs. A very less explored area is fabric dyes. There are yet many fabric dyes which impart color in the cloth and create toxicity in the environment. This toxicity might well be acute, may it be through occupational basis or through consumer basis. We will explore this toxicity in detail in the paper.

There are fabric dyes which are irritant in nature while they are being imparted on the cloth. There are instances where chemical reactions are catalyzed through different chemicals which are toxic during the process of imparting color. Such use can cause intense toxicity to occupational members who are working in the industry or production of colored cloth.

Secondarily, there are also problems with disposal of remaining water. The dye when imparted on the cloth through the process; produces waste water. When this waste water is thrown or disposed in water bodies or in soil; they pollute the soil / water with heavy metals as well as carcinogen compounds. When this water is used as irrigation supply or the same soil is used for agriculture; there is transference of such compounds into the vegetable and other farming products. These products when consumed creates various problems to the human body.

Lastly, when few fabric dyes which absorb UV radiation are worn; they impart their characteristics in intense sunlight. There is also minor exposure to dyes reacting with body sweat and body fluids which can cause trouble with skin. Few colors which come in contact

with urine can cause irritation and such incidences usually occur when harmful dyes are used in manufacturing undergarments.

Hence, we will explore multiple areas where fabric dyes cause toxicity to us in our daily use, along with toxicity towards occupational workers in manufacturing process.

1.2 Fabric Dyeing & Its Extensive Use

Fabric dyes are used in various clothing items and different fibers need different types of dyes to adhere on it. Some dyes are exclusive for cotton and some are exclusive for polyesters and nylons. The characteristics of the fiber gives us the characteristic dye to be used on it. Alongside, there are also different techniques used to fix those dyes on different fabrics. Dyeing is a process of application of pigments on textile and fabric materials for achieving colorfastness. Dye molecules adhere to the fiber creating bonds and these bonds get attached permanently through controlling factors such as temperature, interaction, pH, etc.

Few types of dying which are usually used commercially and are widespread techniques are mentioned below:

1.2.1 **Direct Dyeing**: In this process, no affixing agent is used for putting dyes on the fabric. It is directly applied on the textile material. They can be natural as well as synthetic. This method can be extensively used on cotton fabrics and this is all-purpose dyeing. Since, it mixes with other dyes it cannot be as bright as other dyes. The wash fastness is also not so good but can be improved with treatments with certain agents.

1.2.2 **Stock Dyeing**: It is a process where the fibers are dyed before it is spun. Materials are put into large vats and heated. This gives heather-like effects and weavers more control on wool. It is used on wool and for different shades, different colors are mixed in different proportion.

1.2.3 **Yarn Dyeing**: In this process the dye is applied on the yarn before it is processed into knitted fabrics. This shall give bright colors and applied easily through different agents. [1]

1.2.4 **Skein Dyeing**: This is a process where yarn is dipped into vat dyes. They are loosely dipped where dye is infused into it and further knitted into fashion. It is the oldest and the most expensive method of coloring yarn.

1.2.5 **Garment Dyeing**: This is the process where garments are dyed by placing it into different net bags and immersing it into vat dyes. They remain inside the bag loosely so the dye is infused into it. This method is not suitable for tailored clothing.[2]

1.2.6 **Package Dyeing**: This is a process where yarn is wound on a small spool or tube. This spool or tube is called package and several such packages are fit into dyeing machine at once. This makes a large proportion of fabric dyed very faster; making it reliable and cheap practice.

1.2.7 **Super critical fluid dyeing**: This is a process where carbon dioxide (CO_2) is used as a dyeing medium which collects and recycles itself after every cycle. No drying is required and less energy is required for this method. It is an advance industrial technique used for dyeing.

1.2.8 **Denim**: This is a concept where indigo is used for dyeing purposes. This requires a lot of energy and water and generates a lot of waste product. It is not at all eco-friendly and requires chemical fixation methods.

1.2.9 **Printing**: This is a technique which uses application of pigments and inks on the fabric. This is developed in ancient times and it is one of the oldest form of dyeing. However, it requires a lot of labor and skill. There are 4 main sub-types of printing – Block, Roller, Screen & heat transfer. They are similar to direct dyeing and requires background color dyeing process. This is easily reduced by reducing agents.

1.2.10 **Ultra Sound Technology**: This is a new technology where ultra sound is used by an ultrasonic transducer to apply dye on a fabric or textile kept on a vibrating place through electric generator. This uses lower temperature and low dye. However, it requires few chemical combinations and machinery.

1.2.11 **Disperse Dyeing**: This technique is used in synthetic fibers and it is specially made for nylon materials. This happens at high temperature and it was originally made for cellulose acetate. They ware water insoluble and thus are non-ionic for dying on hydrophobic materials.

1.2.12 **Reactive Dyeing**: This is the most popular technique that is used to dye cotton and cellulose fabrics. This process forms a covalent bond between the fiber and the color and gives greater vibrancy and fastness. The fabric is immersed in dye bath and reactive dyes with water and alkali chemicals are added for fixation. At high temperature and specific pH; it gets fix onto the fabric and shows even color.

1.2.13 **Pigment dyeing**: This process uses pigments rather than dyes and they do not chemically attach with the fibers. This process uses binders through which the pigments are coated on the upper layer of the fabric. They are heat-set onto it.

1.2.14 **Acid Dyeing**: This is a method through which protein fibers are dyed; e.g. wool, silk, nylon, etc. The fabric is immersed in acid bath and heat is applied for fixing the acidic dye onto it. They give good colorfastness and bright colors.

1.2.15 **Tie-Dyeing & Batik**: These processes are techniques from which different patterns are created on fabric and it is a form of resist dyeing. Few portions are tied to resist the dyeing process in Tie-Dyeing and in case of batik dyeing, wax is applied for resisting the fabric on certain portion. This creates intrinsic pattern on it.

1.2.16 **Sulfur Dyeing**: In This process, usually dark shades are produced and fabric is dyed in the bath containing a sulfur dye. The dye is oxidized and it develops color. It is cheap and economical for dark shades. [3]

1.2.17 **Azoic Dyeing**: it is synthesized on the fiber itself. Here, the fabric is treated with di-azoic compound which ensures a coupling movement and reacts on forming an azo dye. This applied dye directly on the fabric and it also gives bright colors and good fastness.

1.2.18 **Indigo Dyeing**: This is an ancient technique used for dyeing textiles and is traditionally used for dyeing denims. The fabric is repeatedly kept into dyeing bath and it is dipped into it and exposed to air alternatively. This manner, the dye is oxidized by the air eventually; creating color.

1.2.19 **Jig Dyeing**: In this method, an open system is set up that takes up two bars on each side. One is higher and another is lower. Every-time goes in and out, dye is picked

and applied on the fabric. The process is repeated for tighter construction. The fabric is watched and not allowed to create crease with every reel.

1.2.20 **Beam Dyeing**: In this process, the dye is allowed to penetrated the fabric by pumping it through the fabric. The fabric is wrapped on a beam having small perforated holes. It is then placed in dye-bath and dye is pumped evenly on it. Machines are used for industrial production for consistent dyeing. [4]

1.2.21 **Bale Dyeing**: This is a low-cost method where fabric is dyed through cold water bath. Imitation chambray is dyed through this method and is a very cheap method to dye cotton materials.

1.2.22 **Burl Or Speck Dyeing**: This is performed on wool and here the colored specks are covered by using special-colored inks in many shades. It is done by hand and no machine is used. [5]

1.2.23 **Chain Dyeing**: This is used in yarns and clothes are usually low in tensile strength. The pieces of clothes are stacked on each other and run through continuous chain of coloring where dyes are infused. This method is suitable for large production.

1.2.24 **Cross Dyeing**: This is one of the widespread methods used for dyeing where variety of colors are used to get effects in one dye bath. It is used to dye fibers with different affinities and usually performed with a blue shade.

1.2.25 **Dope Dyeing**: This method is also called spun dyeing where pigment is bonded in the solution and picked up as the filaments. It is used for cellulose as well as non-cellulose fibers. It is used for bright colors and clear materials.

1.2.26 **Shibori Techniques**: These are different traditional techniques which are modified for dyeing. It uses folding of fabric or wrapping it on poles to produce different patterns in dye bath with every cycle. This produces pattern and customizable shades through multiple dyes. [6]

1.2.27 **Ombre Dip Dye Technique**: This technique is used to produce different gradients and it is most suitable for pillow covers and bedsheets.

1.2.28 **Overdyeing**: This is a technique by layering multiple dyes to produce more depth and variation on the fabric. It is commonly used in yarns and produces a stunning effect.

1.2.29 **Ice Dye**: In this process, ice is used to resist the control color on the fabric. Ice melts and slowly fades and shows color distribution on the fabric. The fabric is covered in ice and mounted. Powdered dye is used onto it and ice is allowed to melt to create dyes patterns.

1.2.30 **Solar Dyeing**: This uses special photosensitive color that is used as dye solution. It activates color when it comes in contact with exposure to sunlight. Fabric is soaked in photosensitive dye solution and exposed to sun. After exposure, the dye is washed and excess dye is removed. [7]

1.3 Dyes & Its Classification / Toxicity.

Dyes were initially made from plant and vegetable extracts in ancient times. Indigo was a prime dye used worldwide and was exported through Asian countries. However, it had tedious and expensive manufacturing process and it needed a lot of labor. European countries came with the idea of synthetic dyes extracted from artificial materials and it soon became famous due to its cheap availability. Modern dyes use various chemical composition which make them toxic. Modern dyes are of many different types:

1.3.1 Azo Dyes: They are the largest class of synthetic dyes and are used extensively in many branches like pharmaceuticals, textile, paper, leather, etc. It is also used in food and pain industry. This is because it shows structural variations and can give us different shades. However, few azoic dyes are banned in many countries of the world like EU, China, Japan, India & Vietnam. US and Australia are still considering them to ban it. This is because it has harmful aromatic amines and are carcinogens. It is possible that exposure to these dyes in presence of its dispersion conditions (may it be extreme sunlight or biological fluids) can emit these amines causing carcinogenic effects on the body. They are classified into Diazo and Tri-Azo respectively. Same dye can give different shads due to its complex structures.

1.3.2 Nitro Dyes: These are multivariate dyes which contain phenol derivatives and at least one nitro group. It may contain it in ortho or para condition. It may also contain aromatic benzene or naphthalene group. It may show moderate fastness.

1.3.3 Indigoid Dyes: These dyes come from indigo chemicals and come from natural as well as synthetic sources. It is one of the most ancient dyes used. Naturally it comes from leaves of Indigofera plant and it is insoluble in water, alcohol and ether. Hence, nitrobenzene, sulfuric acid and other chemicals are used to impart them onto fibers in the dyeing process. It is basic in pH and shows very good color fastness. There are traditional as well as industrial techniques used for dyeing through this dye.

1.3.4 Anthraquinone Dyes: Usually, red-blue color is imparted through this dye and they are obtained through ionic interactions. Electron donor groups or amino / hydroxy groups are used in the dyeing process. However, these dyes can persist in environment as its environmental degradation is not possible for long amount of time. When released in water bodies, it can reduce the penetration of sunlight, effect biological life and sustenance of plants and fishes. It can cause acute toxicity and

microbiological treatment is advised. Although, it is used in various pharmaceuticals and textile industries.

1.3.5 Phthalein Dyes: These are dyes which change color as their pH is adjusted and thus, they fall into organic type of dyes. Theya re a type of synthetic dyes and they are more commonly used in pH indication. They are made from reacting Phthalein anhydride with phenols with a dehydrating agent present in it. It might also contain aryl-compounds and it is also used in titrations except for fabric dyeing. It does not offer much fastness and it is also chronically toxic in nature.

1.3.6 **Triphenyl methane Dyes:** They are synthetic dyes which are used in many industries for printing as well as aquaculture. It is also used in dyeing and printing as coloring agents. It can be degraded biologically and chemical precipitations. It imparts malachite green color and other violet as well as Victoria blue colors. It is used in silk, wool and other fabrics.

1.3.7 Nitroso Dyes: These dyes contain Nitroso group in it and are readily used in rubber, wallpaper and paint industry. They are treated with aromatic hydroxy compounds and added sodium nitrite into acidic medium. They chelate with metal ions and form azo complexes producing different types of pigments. They are affixed on wool, silk and other natural as well as synthetic fibers.

1.3.8 Basic Dyes: These dyes are applied to acrylic, nylon and paper. Theya re also used on polyesters and retarders are used to apply it in boiling conditions. They have poor migration properties and are water soluble in nature. They form bonds with electrostatic attraction with substrate fibers. They are also called cationic dyes and usually localize with ammonium group present in it. It is distributed with delocalized charge on it.

1.3.9 Acid Dyes: These dyes can be easily applied to wool, nylon and cotton. Since the pH lies in acid scale, they are termed as acid dyes. They are applied in acidic conditions in presence of acetic acid and the degree of acid depends on its nature. They include different metal complexes and impart similar differences in color thereby. The bonding happens through Van de Waals forces and gives good color fastness. Usually, sulphonated groups are present in it.

1.3.10 Vat Dyes: They are usually used on cellulose fibers and cotton. They show very good fastness and they are not readily dispersed in water or chlorine. However, for synthetic dyes they are used in alternative purposes as they give pale color. It includes derivatives of indigo and anthraquinone into it. They diffuse with fibers and are water insoluble in nature. [8]

1.3.11 Direct Dyes: These dyes majorly show chromotropic character and are classified as azo, stilbene, dioxazine and other classes like thiazole, quinolone, etc. Theya re easy to apply and have lighter shades with moderate level performance and compatibility. The washing property depends on the fiber substrate on which it has been applied.

1.3.12 Reactive Dyes: These dyes have very high strength and are less expensive than direct dyes. However, it is not easy to obtain them. It shows fastness in extreme conditions but the chlorine fastness is not so good. In chlorinated water, the dyes show less fastness and through daily wash; it fades it color. Since, they are reactive; they form covalent bond with the substrate fibers and it is almost used on wool and nylon. [9]

1.3.13 Sulfur Dyes: They are highest in production rate and it contains variety of colors into it. They include relatively common aromatic compounds and at least have one or more nitro groups attached with it. It may contain benzene, naphthalene, azobenzene, etc. It is used in silk and certain types of leather. They show extreme fastness and ease of application. However, the consumption of sulfur remains high & may have many intermediate aromatic compounds. [10]

1.3.14 Disperse Dyes: These are synthetic colorants and have hydrophobic substrates. They are usually mixed in fabric industries and needs huge amount of water. The amount of wastewater generated is also very large even-though they are insoluble in water showing non-ionic character. It requires aqueous dispersion to get attached to the fibers. Theya re used in polyesters and shows staining effects if the color remains on the fabric. It is affixed through the boiling process.

1.3.15 Solvent Dyes: These dyes are made up of soluble chlorinated hydrocarbons and alcohols. They are insoluble in water but soluble in alcohol or ammonia. Theya re useful to color synthetic materials and gasoline. Primary color in solvent is applied through non-polar flushing technique.

Toxicity in dyes are often unnoticed. There is enough regulation in cosmetic act for hair dyes; however, there are similar chemicals and toxins present in fabric dyes. These fabric dyes are exposed to us daily through undergarments, clothes, bedsheets, pillows, etc. These toxic effluents and chemical merely spread through contact exposure, or through reactions with biological fluids or even through occupational and environmental toxicity. Various heavy metals are found in synthetically treated fabrics and its disposed water. It is thus, important to analyze dyes and need more clarity on its regulated use.

There are various chemicals which are used in textile industry and other fabric dyeing industries which are hazardous to mankind. A few of them widely used are:

1. Formaldehyde
2. Sodium dodecylbenzenesulfonate (SDBS)
3. Alkylphenol ethoxylates (APEOs)
4. Alcohol Ethoxylates (AE)
5. Polyethylene Glycol ethers (PEG Ethers)
6. Alkyl Polyglucosides (APG)
7. Fluorosurfactants
8. Arsenic in Trace amounts in fixation process, etc.

These toxins do not pollute the environment only; they also come in contact with our skin; go into agricultural products and may also cause occupational toxicity to labors working under conditions of extreme conditions. The toxicity of dye and dye-infused chemicals harm the climate, aquatic life, labors and also to those who wear the fabric – by contact exposure.

CHAPTER 2: LITERATURE REVIEW

1. "**Progress towards Circulatory Polyester and cotton Textile**"; by **Siyan Wang & Sonja Salmon** has elaborated the Acid-Alkali and Reflux techniques used for extraction of dyes; has explained in-depth analysis of fabric and dyeing process. They have also described how different fabric materials need different methods of dye fixation and how these different fabrics need selected chemical processes to extract dye from them. They have talked about the efficiency of Acid-Alkali Hot extraction process, Refluxing Process and other laboratory processes. The main agents that they have defined in the process are:
   - Sodium Hydrogen Sulfite,
   - DMSO (Dimethyl Sulfoxide),
   - N-N-Dimethyl Formamidine,
   - DEG (Diethylene Glycol),
   - NMMO (N-methyl morpholine N-oxide),
   - Enzymatic Hydrolysis,
   - Fermentation,
   - Anaerobic Digestion,
   - Acid Hydrolysis,
   - Alcoholysis, etc.

2. "**Biological Color Stripping: A Novel technology for removal of dye from cellulose fibers**"; by **Shahzad Ali Shahid Chatha, Muhammad Asgher, Shaukat Ali & Abdullah Ijah Hussain**; have also mentioned techniques that can extract fabric dyes from the clothes. They have researched on removal of color from clothes. They have talked about biodegradation methods using various microbes and chemical reduction methods. A few bleaching techniques are also mentioned in this article but they disperse color more than they extract it.

3. "**Honorable Discharge: Decolorization of Natural Fabrics**"; by **Karren Brito** mentioned different agents and chemicals such as: Tin Chloride, Jacquard Discharge Paste, Zinc Formaldehyde

Sulfoxylate, Calcium Formaldehyde Sulfoxylate, etc.; which are readily used to discharge colors from different fabric.

4. "**Textile Dyeing: The Step-by-step Guide and Showcase**" by **Kate Broughton,** there are certain hand dyeing methods such as oxidative discharge, using Anti-chlor agents, thickened bleach, through detergents, using fading agents and reductive discharge methods are also mentioned

5. "**Complex Cloth: A Comprehensive Guide to Surface Design**"; by **Jane Dunnewold**. Has also mentioned various types of materials and methods used for dyeing various types of fibers. Acidic dyes are used in certain fabrics and so are basic dyes used in certain fabrics. Dyeing methods also differ due to this variation.

6. "**Textile Preparation and Dyeing**"; by **Choudhary, A. K. R**. have also mentioned types of fibres with their structure and how dye bonds with they essential properties of the system. Cellulose fibres and synthetic fibres are also distinguished and treatment methods are also mentioned. Importance of water softening and sequestering agents have also been mentioned along with water quality and effluent characteristic. Reactive and oxidative processes with surfactants and reductants are also explained in this thesis and dyes are well classified into it by their properties, structure, colours, fastness, etc. Dyeing machines have also been explained along with some ecofriendly methods.

7. "**Textile Science**" by **Gohl, E. P. and Vilensky, L. D** have mentioned complete explanation of fibre properties. It has mentioned subdivision of cellulose fibres as ester-cellulose fibres and some protein fibres. It has also mentioned a lot about chemical finishing needed for proper dyeing of every type of fibre. This edition has enumerated many properties along with its effect and interaction with dyes.

8. **"Watson's Textile Design & Colour"**; by **Grosicki Z** is a very majestic thesis which describes all minute detail about weaving, construction, elements and shades of colour, dyes and effects made into different types of fabrics. It also explains compounds which are known for giving such effects and gives us a very broad idea about fabrics, weaving and dyeing in general.

9. "**Textile Fabric's & Dyes**"; by **Touseef Younas, Noor Tayyaba & Afsheen Ayub, Shaukat Ali** have listed all different types of dyes used for different varieties of fibres made in clothing. This is a go-to literature for short; yet quick idea for understanding the fabric dyes and its utility in no time.

10. "**Classification, properties and application of textile dyes: A review**"; **Benkhaya Said, Sara El Harfi & Ahmed Elharfi** have minutely explained the chemical structure behind all dyes and its interaction in making fixated bonds with the fabric. It is a short review which emphasizes on the chemistry behind different types of fabric dyes and its analogy.

Various methods of dyeing are used commercially and into small retail shops which gives color fixation on clothes and other materials. These methods of dyeing change from region to region; from fabric to fabric and from need to need. This dyeing technology has evolved from ancient times. As evolution has begun; more use of commercially developed synthetic dyes have been increased and natural products are reduced. Many such dyeing process are widely used in India as well as different parts of the world.

Forensic scientists and social agents are eager to test these substances for checking its effect and toxicity on humans. Few dyes impart color but also bring chronic toxicity. There are also chemicals that adhere into fabric in the process of fixation of dyes on it. Now, this again depends on the type of bond a particular dye is forming; polymerization, covalent bond, ionic bond, etc.

Let us look on the extraction process of these chemicals from the fabric. We use different techniques to remove these agents that have bound with the fabric. Some call it Anti-dyeing and few of these processes

are also used in recycling of clothes. Textile fair has recently produced an article defining and suggesting various methods used for separation of dyes from fabric. These include:

- Micro Filtration,
- Ultra Filtration,
- Nano Filtration,
- Reverse Osmosis,
- Electrodialysis,
- Adsorbent Method through Activated Carbon,
- Sugarcane waste recycling,
- Polar Extraction through Peat,
- Ash & Coal Extraction method,
- Silica Gel Method,
- Ozonation,
- Photochemical reduction,
- Oxidative Process,
- Bacterial Decolorization,
- Using Yeast / Algae for the dye separation, etc.

CHAPTER 3: AIM OF THE STUDY & OBJECTIVES

3.1 Aim of the Study

The aim of the study is to find the best suitable reagent and condition for extract of dye from the fabric through hot alkaline Acid-Alkaline Extraction process; Find out toxins and hazardous substances found in these fabric dyes through laboratory processes & to analyze the disposed dye-infused waste water instrumentally.

3.2 Objective

To identify the toxins, present in the fabric dyes; to quantify these toxins through instrumental process (from disposed waste water) & to list the hazardous substances present in these fabric dyes. Our research shall eliminate and expose certain chemicals which might be carcinogenic to humans and be present in our daily worn clothes and apparels. These chemicals may provide bright colors, good wash-fastness but may also cause chronic problems with exposure to biological fluids and other environmental factors. We shall identify and raise concerns about usage of these dyes in fabric use; iff, we found any positive results in our study.

CHAPTER 4: AREAS OF DANGER

4.1 Toxicity – Occupational & Environmental

The field of occupational toxicity developed for finding chemical and biological hazardous exposure at workplace which might bring adverse effects on workers and their family. The exposure to chemicals and raw material used for production of goods might as well be toxic in nature. There are many cases where the finished product might not be toxic in nature but the process of making or manufacturing involves toxic conditions. Occupational toxicologists find these conditions and exposure chemicals which might be a reason for ill-health of the labors working in that environment. These might be complex exposures or direct exposure – in acute or chronic manner. Usually, the chronic exposure happens slowly and one does not notice it until it has reached to a massive stage of toxicity.

The most common form of occupational toxicity when we talk about dyeing industry if water pollution. These dyes are often disposed in nearby water bodies disrupting the water cycle and its harmful effects causes a cycle of toxic exposures. Majorly, the workers working in dyeing industries face toxic agents such as:

- Lead,
- Arsenic,
- Copper,
- Mercury,
- Nickel,
- Cobalt,
- Formaldehyde,
- Toluene,
- Benzene,
- Xylene,
- Nonly-Phenol Ethoxylates,
- P-phenylenediamine,
- Toluene-2,5-diamine, etc.

There are many chemicals which run into water bodies and the same water runs into vegetative production. These chemicals and toxins like chromium, cadmium, lead, etc. aggregate into vegetable and other agricultural products to form bioaccumulation. This again leads to eruption of diseases in long-term by coming it into food chain. Other problems are leaving behind carbon footprints and energy consumptions at large scale. It not only generates waste on large scale but also depletes natural resources. They also give invitation to various problems like:

- Skin irritation,
- Allergies,
- Respiratory problems,
- Coughing,
- Wheezing,
- Breathing disorders,
- Eye & Mucous Membrane Irritation,

- Damage to senses,
- Skin Absorption,
- Carcinogenicity,
- Endocrine Disruption,
- Hormonal Issues, Etc.

4.2 Disposal & Sustainability

When such increasing concerns raise worldwide on the production of fabrics through these synthetic toxic chemical dyes; world is getting evolved in making technology safe. Many measures are to be taken for reducing this damage to humans and environment. Natural Dye alternatives, plant-based dyes, biodegradable dyes and digital printing has been established in many regions. However, economic bulge does not allow these measures to be spread evenly, very fast. Many industries still rely on cheap product and chemical processes to induce dyes on the fabric generating toxicity in the environment. There is an urgent need for regulation and measures which need to be implemented to reduce this damage occurring in the surroundings. This does not only affect humanity but is also affecting aquatic and forest life.

The main problem arrives in disposal is that the dyes which are acidic or basic or reactive in nature; they are highly soluble in water. These dyes discharges are difficult to be removed from water by conventional techniques. They impart color by a substrate due to chromotropic groups into them. The process of fixing it also includes reacting them with various chemicals and effluents. Including aesthetic damage; they also prevent photosynthesis of plants and aquatic weeds beneath the water; disrupting the entire life cycle. It also generates harmful biological mutations and affect growth of plants and forest life.

Another problem are anoxic sediments which get deposited and the aromatic amines which are involved in the process of dyeing. These products take long to degrade and often leads to bioaccumulation. The consequences of this are not very friendly. Many dyes cause bladder cancer and CNS dysfunction on prolong exposure to it (talking about occupational toxicity). Certain chemicals used in these dyeing process causes conjunctivitis, rhinitis, asthma, allergic reactions and contact dermatitis. It forms a conjugate with human serum albumin and the reactive dyes are often a source of Allergic Endocrine Reactions. Moreover, the genotoxicity is another concern that will affect long term hazard to human life. This leads to mutagenic potentiality through various dyes like Azure-B and it changes the structure of DNA and duplex RNA. This occurs by lipid membranes and create cytotoxic effects through intracellular enzymes in nervous system. Many of these dyes are carcinogenic in nature and many cause tumors in different exposed organs.

Many scientists are using Phytoremediation to remove the toxic effects of chemicals through biological and microbial exposure and its life cycle. These microflora and other organisms are used to give remarkable recovery of environmental stress through autotropic bioreactors. Bioremediation and bioaugmentation is also an evolving subject of research for the development of natural cleansers of environmental toxicity.

4.3 Contact Exposure

Many chemical agents used in dyes cause toxicity when exposed to the body. May it be in occupational manner in manufacturing industries (which affect the labors acutely and chronically); may it be in household manner (where washing the clothes and water waste of the dye is exposed to hands of washerman); or may it be to the wearer (when chemicals in dyes react with body fluids like sweat, vaginal fluids, etc.) In any of the manner; the dyes get exposure to the skin and cause toxicity if there are toxins and hazardous elements used in it. It is also possible that when UV light and sunlight is irradiated on it; it released certain fumes and toxins touching the skin on the wearer. However, we need more research to find out how and till what extent does this occur.

There are several heavy metals which leave traces on hands and other exposed regions which might be ingested accidentally or can penetrate the body through any openings or absorption through skin. Since, these are very minor additives; no major effect is seen suddenly. Regular exposure might cause chronic toxicity, carcinogenicity from these chemicals affecting the body. Thus, one needs to explore the dyes and its chemical agents used in fixation; weather they cause harm to the body chronically.

Various chemicals and toxins found in dyes are carcinogenic and many of these cause toxicity to humans. It is neglected because of its trace amounts but regular exposure may cause these negligible effects to get pronounced. They

might become skin irritants and cause long-term toxicity or allergies when one is exposed to it on daily basis. Thus, this field needs more exploration and more research to find the safety of using many different dyes in undergarments as well as sportswear (when more sweat exposure is seen). [11], [12], [13], [14], [15]

4.4 Acute & Chronic Toxicity & Adverse Effects

Acute and chronic toxicity occurs when long-term exposure happens from toxins and hazardous substances present in the dyes. Since it exhibits low toxicity at immediate effect, most of this water waste is disposed in water bodies. This causes harm to aquatic life and agricultural products. Anionic acid and reactive dyes cause skin irritation and respiratory problems. Diverse toxic effect take place even in lower concentration of the dyes and causes effects like nausea, headache, genotoxicity, skin rashes, cancer, etc.

Most of these issues arise from metal oxides present in the dye; which includes:

- Iron Oxides
- Copper Oxides
- Manganese oxides
- Nicle Oxide
- Selenium Oxide
- Zinc Oxide
- Titanium Oxide
- Ruthenium oxide
- Rhodium Oxide
- Tantalum Oxides
- Palladium oxides
- Iridium Oxides
- Osmium Tetroxide
- Platinum Oxides
- Hafnium Oxides
- Rhenium Oxides
- Tungsten Oxide, etc.

These chemicals heighten bioaccumulation significantly. This causes pollution as well as harmful effects to life and living surrounding the region. Diverse industries dispose diversified wastewater and notably cause severity. Dissolved organic matter and other direct dyes also cause various effects and interactions in natural cycles and environment. Precipitation, pollution and disturbances are comprehensive effects caused by the usability of these dyes in everyday life. Many of the above-mentioned oxides also cause cytotoxicity and few cause intense damage to multiple organs of the body. Heavy metals like Arsenic, Copper, Cobalt, Cadmium, Nickel & Chromium affect lungs, kidneys, liver, breasts, bladder, etc. [16]

Over and above, the decomposition and decay of this matter also take long time frame. This dissolved organic matter cause disruption in cycles of life and nature. This causes contamination and thus; one needs to manage the waste getting accumulated and transferred to human life.

Long term exposure potentially leads to allergic reactions, developing allergies, adding toxins to vegetation and agricultural products through irrigation and other harmful health issues. One may develop skin irritation and issues along with repeated exposure may give long-term poisonous effects. These issues can trigger human life directly and indirectly. Disposed toxins may also travel through soil into vegetation and propagate into food cycle causing long term exposure to population living nearby areas. Thus, one needs to address this toxicity with care and concern. [17]

CHAPTER 5: METHODOLOGY

5.1 Methodology

Our approach and method for this research is totally based on selecting local products which are made from average and cheap quality fabrics as well as dyes. This is because the large lump of middle-class people selects these types of garments and are exposed in majority from the population. The main question asked by our research is whether or not any hazardous and toxic substances are present in our daily worn outfits. Especially, those which are in direct exposure and contact with our skin and body fluids (like undergarments). The secondary question lies in weather these toxins are accumulated only in our body or are they accumulated in our environment from frequent washing and disposal of industrial waste-water.

There are many methods of extraction and analysis but our approach will be simple. We would be trying to extract dyes from Hot alkaline Acid-Alkali Extraction Process. This will allow the dyes and chemicals attached to the fabric to get dispersed and extracted in alkaline treated water solvent and the further acid treatment will precipitate those chemicals out of it. For this purpose, we shall test for various reagents and solvent base and choose the best solvent for extraction of dyes from the fabrics. We will conduct the same extraction through different conditions and choose by comparison the best combination for most efficient extraction. Once the extraction is complete, we shall perform additional tests on it to confirm the presence of hazardous substances and identify it.

5.1.1 Research Philosophy

The research philosophy is based on performing qualitative research over fabric dyes attached with the clothes. We would not go for quantitative research at this moment; this is because we are finding the toxins and its causes and not going into the quantification of it. The presence itself shall give us positive result and the absence shall bring us to a negative result. We will start with pragmatic philosophy at first and once we reach our half-way conclusions (by finding substances involve with the dyes); we might go towards positivism approach.

Our fundamental assumption is that dyes and its treatment bring enough toxins on the fabric to bind with it. This binding becomes weaker and weaker with every wash and it readily disposes itself on the skin or waste water. Another assumption is that with every exposure of dyes on body and its interaction with biological fluids; the heavy metals as well as other mixtures or effluents shall react and become toxic. Until this is not proven otherwise; we shall go with this philosophy and perform different research on extracted dyes from fabrics of daily use.

5.1.2 Adopted Method

Our primary research would be qualitative in nature. We shall extract dyes from different fabrics through different reagents and compare to find the best suitable combination from all of those. After finding the suitable method of extraction, we shall extract an ample amount of dye from the fabric. This extracted dye shall be tested through various chemicals for any toxins present into it. Further, instrumental analysis would be performed thereby. The

adopted methods are taken from different literatures but our literature shall compare and contrast different methods of extraction to find the best suitable one.

The extraction performed shall extract all the toxins along with the dye from the fabric into the solvent base. This would further be separated through acidifying the solution. The precipitated substance includes the dye along with additional effluents. Other alternative methods such as reduction and bleaching shall disperse the dye but shall not extract it along with all the toxins present in it. For better result, we are performing hot alkaline extraction with reflux action. The refluxing shall not allow any hazardous extra chemical to fall off into steam or other manner. This is why the flexible method is chosen which could be applied to variety of products and brings us richer understanding about toxicity present in fabrics.

5.1.3 Approach

The present adopted research approach is deductive in nature. We shall deduce finding through qualitative research and perform tests which allows us to build broad idea from pivot testing various fabric dyes. We shall not perform quantification as the amount of toxins are not important for our current research. We need to identify the hazardous agents used in fabrics and present in our daily clothes. Compared to other approaches, our approach is straightforward and easier to deduce our conclusions.

Perspective and meaning-marking methods shall also be performed on the consequences and conclusion part of the research which shall bring deeper insights about the usage and effects of these dyes in our daily life. Many researches have been performed in extraction of dyes; all have been found to with positive results but the yield varies. Our research cites the yield of each extraction method and reagent used simultaneously. Furthermore, we shall identify and test for heavy metals and hazardous substances one by one; along with instrumental analysis of our secondary sample – the disposed waste water.

5.1.4 Design

Our research design is based on correlational & Quasi-Experimental research design. The qualitative research has been reported descriptively with proper inferences. A sequential exploratory format has been chosen inform of tables to list all the extraction methods and conditions whereas the conclusions are based on explanatory format.

The research focuses on finding the best method through trial and error and using the concluded method for sampling as well as conditioning of the dye. The final result shall either be positive or negative. Positive results conclude having hazardous substances alongside the list of substances found through our qualitative tests. Negative results eliminate the possibility of having toxic substances in fabric dyes once it has adhered onto the fabric.

5.1.5 Study Population & Sampling

Our sample population consists of different local brands of undergarments (since they are the first to interact with body fluids) as well as stretchable pants and other fabrics used in day-to-day life. A general population of average condition fabric is chosen as to get an idea of what the general localized people are exposed to. Heavy brands are only chosen by few individuals comparatively. Thus, samples are chosen from local stores and vendors of Gujarat, India.

We have taken 2 male undergarment and 2 female undergarments. Along with this, we have taken stretchable pants of females and Bermudas of male. We also took samples from a Jeans Denim as well as socks. We have chosen these samples as it is quite exposed to major population strands in India. These garments were tested with different reagents and conditions and different number of dyes were extracted out from them. The extracted dyes were then collected for further analysis and inference.

5.1.6 Data Collection & Analysis

Samples were collected from vendors at different dates and different range of garments were brought. The date and time and price of the garments are also mentioned alongside the research in the extraction analysis section. The quantity of garments taken were roughly of different types which were divided by genders (male & female).

Once, the dyes were extracted and analysis was performed chemically and instrumentally; the sequence of extraction along with chemical reactions and inferences were derived and noted side-by-side. All the data collection information and analysis is mentioned in tabular form in upcoming section. Different efficacy of data; along with its comparison has been shown too. ISI marked tools were used in the collection and analysis of samples for getting precise readings. Photographs are also added.

Content analysis is narrated through structural approach for easier understanding of the complex experiments performed in the sequence. Sequential experiments were performed and compared side-by-side to get results of desired form. Statistical study was performed on percentage of extraction and further classification study was performed on toxins and hazardous substances found in the dyes. Overall thematic study was performed on interpretive studies over analyzed content.

5.1.7 Ethical Considerations

No brand names are added in the research and no photos are taken showing the brand name, symbol, logo, identity of any type. The samples taken are random from random shops around the city and of random prices and no specificity is seen in choosing the sample. All procedures are performed equally on all samples and no biased outcome has been generated. The undergarments are taken from different local vendors which are of local manufacturers

and sold locally. No specific brand is targeted or tested without any consent. The research has been many purely anonymously with random samples chosen.

The potential risk of ruining the reputation has been taken care off and no names or identities are used. Moreover, the process performed are comprehensive and the outcome are mentioned with clarity. All results are tabularly mentioned and photos are also added for reference. A comprehensive information for each reaction as well as all results have been mentioned with proper details, dates and outcome.

Proper documentation has been performed and appropriate methods are used for testing as well as extracting dyes form the sample. The chosen reagents have been priorly tested in few research and a

comparative analysis has been performed for all reagents and the dye. If different dyes are used as samples; it is mentioned in the column as well.

Privacy and anonymity are also taken into consideration as well as confidentiality is also kept for the data collected from different sources. The data and samples are stored in safe conditions and no harm was caused during the process and reaction. A minimum physical as well as psychological harm concept was chosen and all reactions / procedures are justified with results and inferences in the next section thereby. Negative results are also mentioned under 'Negative Result' heading and inferences for each have been written thereby.

No deception and bias are used in the research and honesty policy is applied overall. No misleading content is practiced as well as reported. Objectivity in data analysis and interpretation has also been free from personal assumptions and personal biases. Proper citation has been done wherever possible and plagiarism is also avoided. In all places where intellectual property rights and copyrighted content is mentioned; it is cited as well as given honor in references.

Data retention, sharing and security has been taken into consideration.

5.1.8 Limitations & Delimitations

Limitations: Few factors that were outside the control of the researcher which tend to be the limitation of the research are mentioned below:

- Resource Limitation: The resources were taken from the local vendors and local shops of one region of Rajkot, Gujarat, Bharat. Thus, locally used materials are targeted. However, selection of different vendors has given us variety and different range products also gave us variety in the products.
- Time constraint: Since, there is a time constraint for submission of the research, no long instrumental techniques are further used in the analysis of extracted dye. However, we have tried to perform comparative TLC as well as Heavy Metal Analysis to ensure fruitful results.
- Sample Size: The sample size is constrained up to 10 samples only. Although, different materials and different vendors and different local brands are chosen for objectivity and generalization purpose.
- Environmental: We have tried to minimize the influence of environment on the research. Although, few dyes readily change in harsh temperatures and shed of color. However, we have taken this in consideration.

Delimitations: The boundary of the scope of the research intentionally kept for the study are mentioned below:

- Specificity has been maintained with selection of objective materials and garments from different areas and vendors. Clothes of different age groups are chosen, may it be children, adults and old aged. This gives us a generalized broad idea.
- We have narrowed down our research to heavy metal and toxins into the dye analysis. This has brought us to closer look on selective toxins and gave us an idea of hazardous materials present in dyes extracted.
- Specific time frame is chosen to avoid environmental degradation and all research has been performed one-by-one on the same week to avoid contamination.
- We have chosen Borosil© equipment which have high precision and does not give us a vague idea.
- Specific methods and reactions are chosen which are verified through early research practices.

5.2 Primary Sample Extraction & Analysis

5.2.1 Extraction through Decoction

Primary samples are extracted from fiber samples from different clothes. Acid-Alkali extraction is used through decoction, distillation and microwave heating. The fabric dyes used in undergarments are alkali soluble dyes. Hence, these dyes are easily extracted by dissolving it into basic solutions at high temperature. Thus, we are using distilled water in alkali conditions (by presence of NaOH) to disintegrate the dye from the cloth. Sodium hydrogen sulfite is also added to catalyze this reaction and allow the molecules of dye to disintegrate the bonds. However, this happens at high temperature conditions (reaching to its boiling point). Once, dyes are dissolved in this solution; the cloth is removed and the solutions is separated. This solution is alkaline in nature. Thus, the dye is soluble into it. When this solution is brought to acidic condition, by adding an acid reagent (usually HCl); the dye is precipitated into it. The precipitate is then filtered using the filtration apparatus using Whatman Filter Paper Grade 42. This precipitate is then allowed to dry using oven at 50℃ for 5 minutes. Further, it is scrapped out and collected for analysis. Extraction of different samples are given further ahead.

Few Samples Before They Were Torn Apart

5.2.2 Sample Extraction

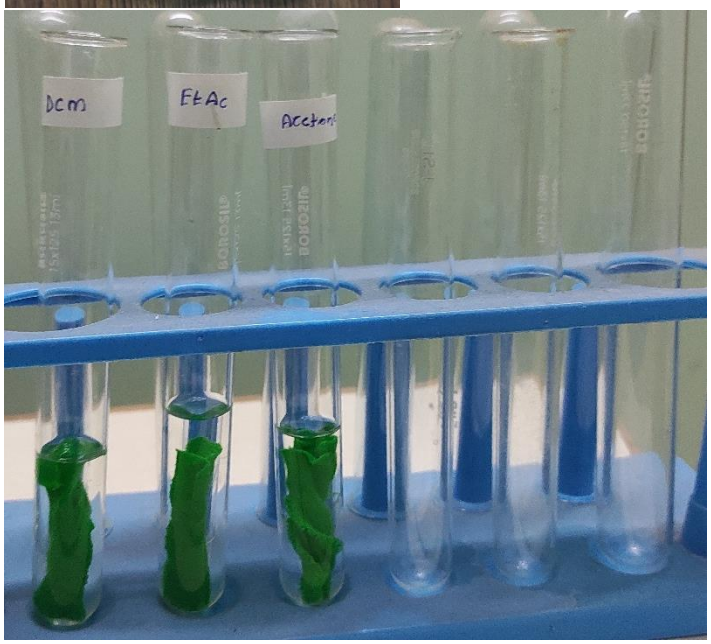
There are few different samples taken from different garments. Undergarments are chosen because they are usually in contact with sweat and other biological fluids which might react with the toxic components in the fabric dyes. The samples are mentioned below:

Children Underwear	
BRIGHT ORANGE	**BRIGHT GREEN**
The undergarment was made from cotton and semi-synthetic material. It did not contain any polymer or plastic fiber. It was not printed and had no adhered logo on it. It was bright orange in color and the tag said it was wash friendly and washing machine friendly.	The undergarment was made from cotton and semi-synthetic material. It did not contain any polymer or plastic fiber. It was not printed and had no adhered logo on it. It was bright green in color and the tag said it was wash friendly and washing machine friendly.
Purchase: 15-01-2025 **Vendor**: Local Vendor in Local Market **Price:** Rs. 150	**Purchase**: 15-01-2025 **Vendor**: Local Vendor in Local Market **Price:** Rs. 150
Dated: **21-01-2025** Pivot Testing was performed by immersing a small piece of cloth in different solvents to check if the dye was dispersing into it. A piece of cloth was torn and was immersed in Acetic Acid, Hydrogen Peroxide (30%), Sodium Hypo-chloride and Water. It was kept overnight and there was no dispersion of dye in any organic solvent except sodium hypo-	Dated: **21-01-2025** Pivot Testing was performed by immersing a small piece of cloth in different solvents to check if the dye was dispersing into it. A piece of cloth was torn and was immersed in Acetone, Isopropanol, Dichloromethane and water. It was kept overnight and there was no dispersion of dye in any organic

chloride. Sodium Hypo-Chloride was able to discolor the fabric but was not able to extract the dye in the solvent. The solvent evaporated and the dye remain unchanged.	solvent. The solvent evaporated and the dye remain unchanged.
Dated: **22-01-2025** The entire undergarment was torn into small pieces to increase the surface area. A solution was prepared of 480ml distilled water with 10g sodium hydroxide and 12g sodium hydrogen sulfite, 3g 30% hydrogen peroxide, 3g sodium percarbonate & 3g Sodium perborate. It was stirred until all the contents had dissolved into it. It was taken into a beaker and heated on a magnetic stirring machine with induction coil. It was brought on boiling temperature and boiled till the water evaporated and 200ml was remaining. The dye had started to disperse in the basic condition. To increase the dispersion, some more sodium hydrogen sulfite was added and the water level was again increased till 300ml. It was again allowed to get evaporated and more dye had been dispersed. Finally, the water level reached around 200ml and it was taken off. The cloth was yet not fully white and some amount of dye was again left on it. Hence, a second decoction and extraction were performed using the same method with same cloth with the same solvent. The process was repeated in the previous manner; but another reagent sodium metabisulfite was added this time (10g). Moreover, 1 tablespoon of glycerin was added as catalyst. The first run took around 1 hour and the second run were around 2 hours. Sodium metabisulfite was able to remove more dye than sodium hydrogen sulfite. Although, the color of dye became more intense in the water and the fabric faded to pale white in shade. The water with dye dispersed into it was taken and collected in another reagent glass bottle and the cloth was kept in another jar.	Dated: **23-01-2025** The entire undergarment was torn into small pieces to increase the surface area. A solution was prepared of 400ml distilled water with 6g of sodium hydroxide and 8g of sodium hydrogen sulfite. It was stirred until all the contents had dissolved into it. It was taken into a beaker and heated on a magnetic stirring machine with induction coil. It was brought on boiling temperature and boiled till the water evaporated and 200ml was remaining. The dye had started to disperse in the basic condition. To increase the dispersion, some more sodium hydrogen sulfite was added and the water level was again increased till 300ml. It was again allowed to get evaporated and more dye had been dispersed. Finally, the water level reached around 150ml and it was taken off. The cloth was yet not fully white and some amount of dye was again left on it. Hence, a second decoction and extraction were performed using the same method with same cloth but fresh solvent. The process was repeated in the previous manner. The first run took around 3 hours and the second run were around 4 hours. After double hot alkaline extraction, the dye was separated but the fabric still showed dull shade of lighter bluish color. Although, most of the color was extracted in water. The water with dye dispersed into it was taken and collected in another reagent glass bottle and the cloth was kept in another jar.
Dated: **25-01-2025** The collected water with dispersed dye was treated with few drops of dilute hydrochloric acid but no precipitate was found. Hydrochloric Acid was oxidizing the solution rather than acidifying it. Thus, 50% Dilute Sulfuric Acid was added into it until dye precipitate was observed. The alkaline conditions were diminished and dye precipitated out in acidic condition.	2nd Extract Dated: **25-01-2025** The collected water with dispersed dye was treated with 50% Dilute Hydrochloric Acid was added into it until dye precipitate was observed. The alkaline conditions were diminished and dye precipitated out in acidic condition. This mixture was allowed to get filtered through filtration apparatus by Whatman Filter Paper (Grade

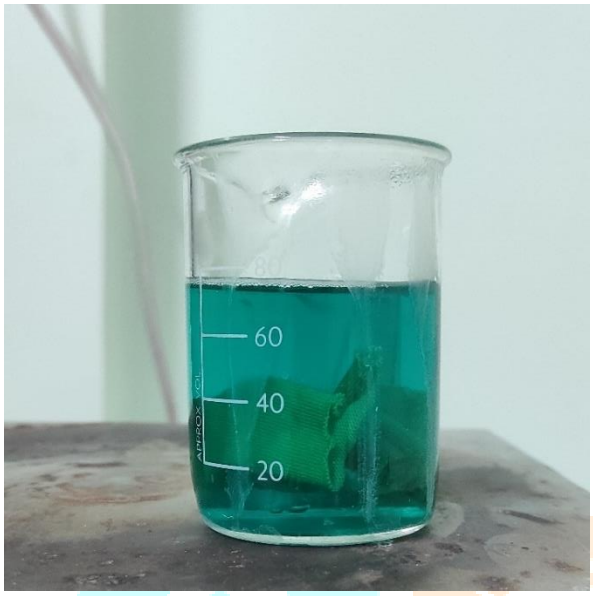
This mixture was allowed to get filtered through filtration apparatus by Whatman Filter Paper (Grade 42). The precipitate was taken into a glass Petri dish and was dehydrated in oven at 50℃ for 2 minutes. The dehydrated content was then scrapped out and collected as the extracted dye. The weight was approximately: **5g**	42). The precipitate was taken into a glass Petri dish and was dehydrated in oven at 50℃ for 2 minutes. The dehydrated content was then scrapped out and collected as the extracted dye. The weight was approximately: **3g** 1st Extract Dated: **25-01-2025** The collected water with dispersed dye was treated with 50% Dilute Hydrochloric Acid was added into it until dye precipitate was observed. The alkaline conditions were diminished and dye precipitated out in acidic condition. This mixture was allowed to get filtered through filtration apparatus by Whatman Filter Paper (Grade 42). The precipitate was taken into a glass Petri dish and was dehydrated in oven at 50℃ for 2 minutes. The dehydrated content was then scrapped out and collected as the extracted dye. The weight was approximately: **3g**

Children's Undergarment : **BRIGHT GREEN**



Pivot Testing On:
Dichloromethane, Ethyl Acetate & Acetone

Torn pieces heated with reagents and solvent to extract the dye while rigourous stirring.



Adding **Organic Solvent** &
Seperating the Organic Phase
Through **Seperating Funnel**


BOROSIL®

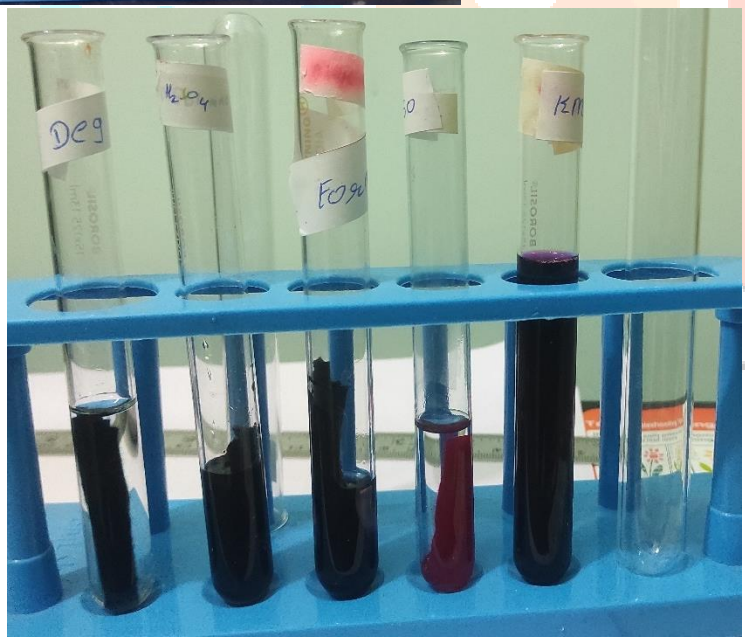
Using **Whatman Filter Paper No.42** To Filter
To Filter The Acidified Precipitate.

Female Stretchable Pants	
BRIGHT PINK - 1	**BRIGHT PINK - 2**
The garment was made from cotton and semi-synthetic material. It did not contain any polymer or plastic fiber. It was not printed and had no adhered logo on it. It was bright Pink in color and the tag said it was wash friendly and washing machine friendly.	The garment was made from cotton and semi-synthetic material. It did not contain any polymer or plastic fiber. It was not printed and had no adhered logo on it. It was bright Pink in color and the tag said it was wash friendly and washing machine friendly.
Purchase: 12-02-2025 **Vendor**: Local Vendor in Local Market **Price:** Rs. 475	**Purchase**: 12-02-2025 **Vendor**: Local Vendor in Local Market **Price:** Rs. 475

Dated: **13-02-2025** Pivot Testing was performed by immersing a small piece of cloth in different solvents to check if the dye was dispersing into it. A piece of cloth was torn and was immersed in N-N Formamidine, Hydrochloric Acid, and Calcium Percarbonate. It was kept overnight and there was dispersion of dye in N-N Formamidine as well as Hydrochloric Acid. Hydrochloric Acid Damaged the fibers whereas the prior one just extracted the dye. Calcium Percarbonate gave bleaching effect and the dye was faded but not extracted.	Dated: **13-02-2025** Pivot Testing was performed by immersing a small piece of cloth in different solvents to check if the dye was dispersing into it. A piece of cloth was torn and was immersed in N-N Formamidine, Hydrochloric Acid, and Calcium Percarbonate. It was kept overnight and there was dispersion of dye in N-N Formamidine as well as Hydrochloric Acid. Hydrochloric Acid Damaged the fibers whereas the prior one just extracted the dye. Calcium Percarbonate gave bleaching effect and the dye was faded but not extracted.
Dated: **14-02-2025** The entire undergarment was torn into small pieces to increase the surface area. A solution was prepared of 400ml distilled water with 10ml of ammonium hydroxide and 8g of sodium Dithionite. It was stirred until all the contents had dissolved into it. It was taken into a beaker and heated on a magnetic stirring machine with induction coil. It was brought on boiling temperature and boiled till the water evaporated and 200ml was remaining. The dye had started to disperse in the basic condition. To increase the dispersion, some sodium carbonate was added. It was again allowed to get evaporated and no more dye had been dispersed. Finally, the water level reached around 150ml and it was taken off. The cloth was yet not fully white and dye was again left on it. Hence, a second decoction and extraction were performed using the same method with same cloth but fresh solvent. The process was repeated in the previous manner but doubling the amount of Sodium Dithionite. The first run took around 3 hours and the second run was the same. The water with dye dispersed into it was taken and collected in another reagent glass bottle and the cloth was kept in another jar. The color change was fast but the overall yield was very less.	Dated: **14-02-2025** The entire undergarment was torn into small pieces to increase the surface area. A solution was prepared of 320ml distilled water with 10g of sodium hydroxide and 8g of sodium metabisulfite. It was stirred until all the contents had dissolved into it. It was taken into a beaker and heated on a magnetic stirring machine with induction coil. It was brought on boiling temperature and boiled till the water evaporated and 150ml was remaining. The dye had started to disperse in the basic condition very quickly. To increase the dispersion, some sodium carbonate was added. It was again allowed to get evaporated and no more dye had been dispersed. Finally, the water level reached around 150ml and it was taken off. The cloth was yet not fully white and dye was again left on it. Hence, a second decoction and extraction were performed using the same method with same cloth but fresh solvent. The process was repeated in the previous manner but doubling the amount of Sodium metabisulfite. The first run took around 3 hours and the second run was the same. The water with dye dispersed into it was taken and collected in another reagent glass bottle and the cloth was kept in another jar.
Dated: **21-02-2025** The collected water with dispersed dye was treated with few drops of dilute hydrochloric acid but no precipitate was found. Hydrochloric Acid was oxidizing the solution rather than acidifying it. Thus, 50% Dilute Sulfuric Acid was added into it.	Dated: **21-02-2025** The collected water with dispersed dye was treated with few drops of dilute hydrochloric acid but no precipitate was found. Hydrochloric Acid was oxidizing the solution rather than acidifying it. Thus, 50% Dilute Sulfuric Acid was added into it.

The alkaline conditions were diminished but the dye was not precipitated out clearly. This mixture was allowed to get filtered through filtration apparatus by Whatman Filter Paper (Grade 42). **No precipitate was found & dye extraction was failed.**	The alkaline conditions were diminished but the dye was not precipitated out clearly. This mixture was allowed to get filtered through filtration apparatus by Whatman Filter Paper (Grade 42). **No precipitate was found & dye extraction was failed.**

Pivot Testing Performed with **Hydrogen Peroxide, Acetic Acid & Sodium Hypochloride.**



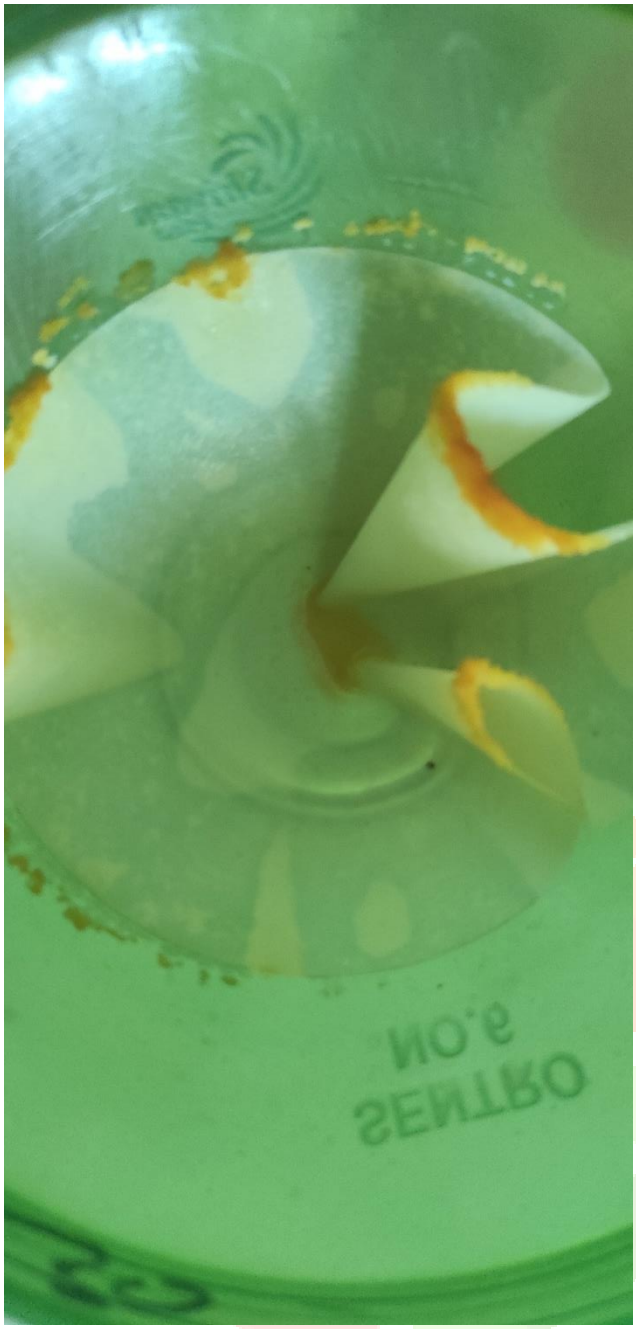
Pivot Testing Performed On: **Diethylene Glycol, Sulfuric Acid, Formic Acid, Potassium Permanganate & Isopropanol.**

Female Stretchable Pants	
BABY PINK - 1	**BABY PINK - 2**
The garment was made from cotton and semi-synthetic material. It did not contain any polymer or plastic fiber. It was not printed and had no adhered logo on it. It was baby Pink in color and the tag said it was wash friendly and washing machine friendly.	The garment was made from cotton and semi-synthetic material. It did not contain any polymer or plastic fiber. It was not printed and had no adhered logo on it. It was baby Pink in color and the tag said it was wash friendly and washing machine friendly.

Purchase: 20-02-2025 **Vendor**: Local Vendor in Local Market **Price:** Rs. 300	**Purchase**: 20-02-2025 **Vendor**: Local Vendor in Local Market **Price:** Rs. 300
Dated: **22-02-2025** Pivot Testing was performed by immersing a small piece of cloth in different solvents to check if the dye was dispersing into it. A piece of cloth was torn and was immersed in N-N Formamidine, Hydrochloric Acid, and Calcium Percarbonate. It was kept overnight and there was dispersion of dye in N-N Formamidine as well as Hydrochloric Acid. Hydrochloric Acid Damaged the fibers whereas the prior one just extracted the dye. Calcium Percarbonate gave bleaching effect and the dye was faded but not extracted.	Dated: **22-02-2025** Pivot Testing was performed by immersing a small piece of cloth in different solvents to check if the dye was dispersing into it. A piece of cloth was torn and was immersed in N-N Formamidine, Hydrochloric Acid, and Calcium Percarbonate. It was kept overnight and there was dispersion of dye in N-N Formamidine as well as Hydrochloric Acid. Hydrochloric Acid Damaged the fibers whereas the prior one just extracted the dye. Calcium Percarbonate gave bleaching effect and the dye was faded but not extracted.
Dated: **23-02-2025** The entire undergarment was torn into small pieces to increase the surface area. A solution was prepared of 320ml distilled water with 10g of sodium hydroxide and 8g of sodium metabisulfite & 10g Sodium Carbonate. It was stirred until all the contents had dissolved into it. It was taken into a beaker and heated on a magnetic stirring machine with induction coil. It was brought on boiling temperature and boiled till the water evaporated and 150ml was remaining. The dye had started to disperse in the basic condition very quickly. To increase the dispersion, some more sodium metabisulfite was added. It was again allowed to get evaporated and no more dye had been dispersed. Finally, the water level reached around 150ml and it was taken off. The cloth was yet not fully white and dye was again left on it. Hence, a second decoction and extraction were performed using the same method with same cloth but fresh solvent. The process was repeated in the previous manner but doubling the amount of Sodium Metabisulfite. The first run took around 3 hours and the second run was 2 hours. The water with dye dispersed into it was taken and collected in another reagent glass bottle and the cloth was kept in another jar. The color change was fast but the overall yield was very less.	Dated: **23-02-2025** The entire undergarment was torn into small pieces to increase the surface area. A solution was prepared of 320ml distilled water with 10g of sodium hydroxide and 8g of sodium metabisulfite, 10ml {30%} Hydrogen Peroxide & 10g Sodium Carbonate. It was stirred until all the contents had dissolved into it. It was taken into a beaker and heated on a magnetic stirring machine with induction coil. It was brought on boiling temperature and boiled till the water evaporated and 150ml was remaining. The dye had started to disperse in the basic condition very quickly. To increase the dispersion, some sodium metabisulfite was added. It was again allowed to get evaporated and no more dye had been dispersed. Finally, the water level reached around 150ml and it was taken off. The cloth was yet not fully white and dye was again left on it. Hence, a second decoction and extraction were performed using the same method with same cloth but fresh solvent. This time 1 tablespoon of glycerin was added as catalyst. The process was repeated in the previous manner but doubling the amount of Sodium metabisulfite. The first run took around 3 hours and the second run was 1 hour. The water with dye dispersed into it was taken and collected in another reagent glass bottle and the cloth was kept in another jar.

Dated: **25-02-2025** The collected water with dispersed dye was treated with few drops of dilute hydrochloric acid but no precipitate was found. Hydrochloric Acid was oxidizing the solution rather than acidifying it. Thus, 50% Dilute Sulfuric Acid was added into it. The alkaline conditions were diminished but the dye was not precipitated out clearly. This mixture was allowed to get filtered through filtration apparatus by Whatman Filter Paper (Grade 42). **No precipitate was found & dye extraction was failed.**	Dated: **25-02-2025** The collected water with dispersed dye was treated with few drops of dilute hydrochloric acid but no precipitate was found. Hydrochloric Acid was oxidizing the solution rather than acidifying it. Thus, 50% Dilute Sulfuric Acid was added into it. The alkaline conditions were diminished but the dye was not precipitated out clearly. This mixture was allowed to get filtered through filtration apparatus by Whatman Filter Paper (Grade 42). **No precipitate was found & dye extraction was failed.**

Negative Result On Extraction Of Dye Of Pink Color.



When the solution containing dye was acidified the color changed to yellow with dispersed particles in the solution. However, when the same was allowed to filter through the filter paper; there was no residue found on the filter paper and no precipitate was formed. Although the reagents worked on cotton clothing, it did not work on the synthetic polyester garments and its dyes.

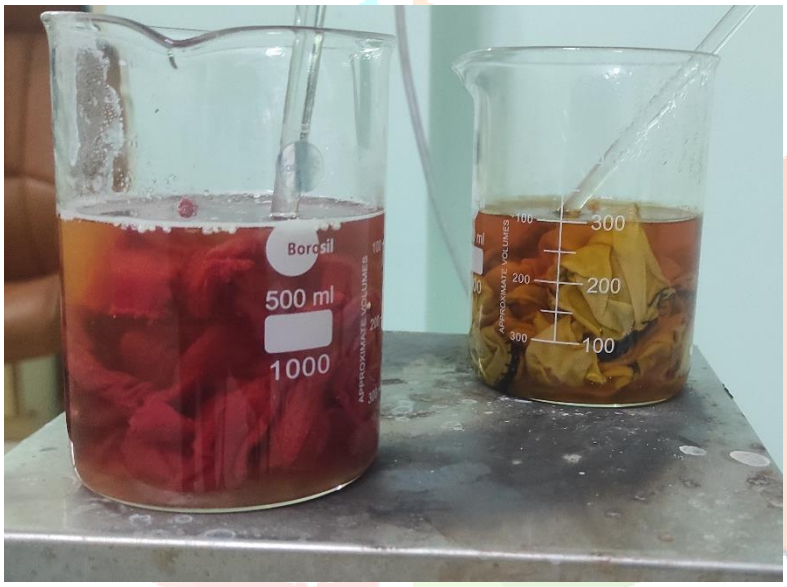
Adult Underwear Male	
DULL RED	**DULL GREEN**
The undergarment was made from cotton and semi-synthetic material. It did not contain any polymer or plastic fiber. It was not printed and had no adhered logo on it. It was dull Red in color and the tag said it was wash friendly and washing machine friendly.	The undergarment was made from cotton and semi-synthetic material. It did not contain any polymer or plastic fiber. It was not printed and had no adhered logo on it. It was bright green in color and the tag

	said it was wash friendly and washing machine friendly.
Purchase: 30-01-2025 **Vendor**: Local Vendor in Local Market **Price:** Rs. 250	**Purchase**: 30-01-2025 **Vendor**: Local Vendor in Local Market **Price:** Rs. 250
Dated: **01-02-2025** Pivot Testing was performed by immersing a small piece of cloth in different solvents to check if the dye was dispersing into it. A piece of cloth was torn and was immersed in Acetone, Isopropanol, Dichloromethane and water. It was kept overnight and there was no dispersion of dye in any organic solvent. The solvent evaporated and the dye remain unchanged.	Dated: **01-02-2025** Pivot Testing was performed by immersing a small piece of cloth in different solvents to check if the dye was dispersing into it. A piece of cloth was torn and was immersed in Acetone, Isopropanol, Dichloromethane and water. It was kept overnight and there was no dispersion of dye in any organic solvent. The solvent evaporated and the dye remain unchanged.
Dated: **03-02-2025** The entire undergarment was torn into small pieces to increase the surface area. A solution was prepared of 400ml distilled water with 6g of sodium hydroxide and 8g of sodium hydrogen sulfite. It was stirred until all the contents had dissolved into it. It was taken into a beaker and heated on a magnetic stirring machine with induction coil. It was brought on boiling temperature and boiled till the water evaporated and 200ml was remaining. The dye had started to disperse in the basic condition. To increase the dispersion, some more sodium hydrogen sulfite was added and the water level was again increased till 300ml. It was again allowed to get evaporated and more dye had been dispersed. Finally, the water level reached around 150ml and it was taken off. The cloth was yet not fully white and some amount of dye was again left on it. Hence, a second decoction and extraction were performed using the same method with same cloth but fresh solvent. The process was repeated in the previous manner but doubling the amount of Sodium Hydrogen Sulfite. The first run took around 3 hours and the second run were around 4 hours. The water with dye dispersed into it was taken and collected in another reagent glass bottle and the cloth was kept in another jar.	Dated: **03-02-2025** The entire undergarment was torn into small pieces to increase the surface area. A solution was prepared of 500ml distilled water with 10g of sodium hydroxide, 3ml Hydrogen Peroxide (30%), 10g of Soda Ash and 12g of sodium hydrogen sulfite. It was stirred until all the contents had dissolved into it. It was taken into a beaker and heated on a magnetic stirring machine with induction coil. It was brought on boiling temperature and boiled till the water evaporated and 250ml was remaining. The dye had started to disperse in the basic condition. To increase the dispersion, some more sodium hydrogen sulfite was added and the water level was again increased till 300ml. It was again allowed to get evaporated and more dye had been dispersed. Finally, the water level reached around 250ml and it was taken off. The cloth was yet not fully white and some amount of dye was again left on it. Hence, a second decoction and extraction were performed using the same method with same cloth with fresh solvent but using sodium dithionite instead of sodium hydrogen sulfite. The process was repeated in the previous manner. The first run took around 3 hours and the second run were around 2 hours. The water with dye dispersed into it was taken and collected in another reagent glass bottle and the cloth was kept in another jar.

Dated: **05-02-2025** The collected water with dispersed dye was treated with few drops of 50% Dilute Sulfuric Acid but no precipitate was found. Sulfuric Acid was oxidizing the solution rather than acidifying it. Thus, 50% Dilute Hydrochloric Acid was added into it until dye precipitate was observed. The alkaline conditions were diminished and dye precipitated out in acidic condition. This mixture was allowed to get filtered through filtration apparatus by Whatman Filter Paper (Grade 42). The precipitate was taken into a glass Petri dish and was dehydrated in oven at 50℃ for 2 minutes. The dehydrated content was then scrapped out and collected as the extracted dye. The weight was approximately: **15g**	Dated: **05-02-2025** The collected water with dispersed dye was treated with few drops of 50% Dilute Sulfuric Acid but no precipitate was found. Sulfuric Acid was oxidizing the solution rather than acidifying it. Thus, 50% Dilute Hydrochloric Acid was added into it until dye precipitate was observed. The alkaline conditions were diminished and dye precipitated out in acidic condition. This mixture was allowed to get filtered through filtration apparatus by Whatman Filter Paper (Grade 42). The precipitate was taken into a glass Petri dish and was dehydrated in oven at 50℃ for 2 minutes. The dehydrated content was then scrapped out and collected as the extracted dye. The weight was approximately: **7g**

Comparative Extraction With Same Material and Different Reagents.
Beaker 2 showed faster extraction of dye than 1 and 3.

A Few Minutes Later… The Color Started To Fade Away



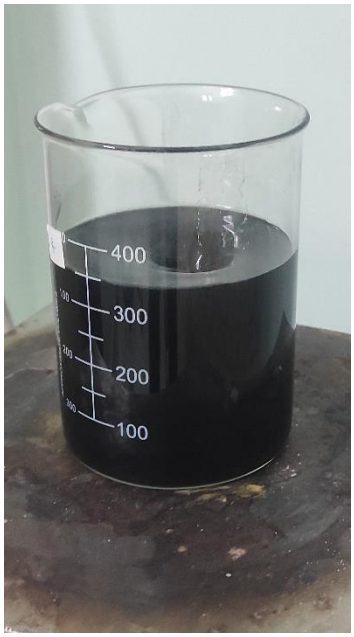
Double Extraction: The second extraction on same solvent with Beaker 1 & with new solvent in Beaker 2. The primary Solvent was divided equally Into both the beakers with equal division of torn garment.

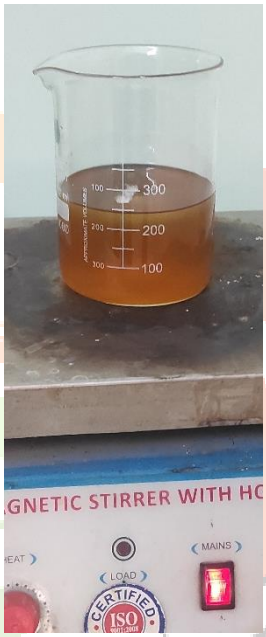
Adult Underwear Female	
BLACK - 2	**BLACK - 1**
The undergarment was made from cotton and semi-synthetic material. It did not contain any polymer or plastic fiber. It was not printed and had no adhered logo on it. It was dark Black in color and the tag said it was wash friendly and washing machine friendly.	The undergarment was made from cotton and semi-synthetic material. It did not contain any polymer or plastic fiber. It was not printed and had no adhered logo on it. It was dark black in color and the tag said it was wash friendly and washing machine friendly.
Purchase: 10-02-2025 **Vendor**: Local Vendor in Local Market **Price:**	**Purchase**: 10-02-2025 **Vendor**: Local Vendor in Local Market **Price:**

Rs. 180	Rs. 180
Dated: **12-02-2025** Pivot Testing was performed by immersing a small piece of cloth in different solvents to check if the dye was dispersing into it. A piece of cloth was torn and was immersed in Potassium Permanganate, Isopropanol, Dichloromethane and water. It was kept overnight and there was no dispersion of dye in any organic solvent. The solvent evaporated and the dye remain unchanged.	Dated: **12-02-2025** Pivot Testing was performed by immersing a small piece of cloth in different solvents to check if the dye was dispersing into it. A piece of cloth was torn and was immersed in Sulfuric Acid, Formic Acid & Diethylene Glycol. It was kept overnight and there was dispersion of dye seen in formic acid as well as in sulfuric acid. Sulfuric acid precipitated out the dye faster damaging the fabric but formic acid brought least damage. Rest showed no dispersion. The solvent evaporated and the dye remain unchanged.
Dated: **14-02-2025** The entire undergarment was torn into small pieces to increase the surface area. A solution was prepared of 480ml distilled water with 10g of sodium sulfide and 6ml of ammonium hydroxide (aq). It was stirred until all the contents had dissolved into it. The solution got pale yellow in color. It was taken into a beaker and heated on a magnetic stirring machine with induction coil. It was brought on boiling temperature and boiled till the water evaporated and 200ml was remaining. The dye had started to disperse in the basic condition even before the heating started. To increase the dispersion, some more sodium formaldehyde Sulfoxylate was added and the water level was again increased till 300ml. It was again allowed to get evaporated and more dye had been dispersed. Finally, the water level reached around 150ml and it was taken off. The cloth was yet not fully white and some amount of dye was again left on it. The water with dye dispersed into it was taken and collected in another reagent glass bottle and the cloth was kept in another jar. The yield was just 50% in this case.	Dated: **14-02-2025** The entire undergarment was torn into small pieces to increase the surface area. A solution was prepared of 320ml distilled water with 10g of sodium hydroxide and 10g of Soda Ash with 10g of Sodium Formaldehyde Sulfoxide (SFS). It was stirred until all the contents had dissolved into it. It was taken into a beaker and heated on a magnetic stirring machine with induction coil. It was brought on boiling temperature and boiled till the water evaporated and 150ml was remaining. The dye had started to disperse very quickly in the basic condition. To increase the dispersion, some sodium dithionite was added and the water level was again increased till 300ml. It was again allowed to get evaporated and more dye had been dispersed. Finally, the water level reached around 150ml and it was taken off. The cloth was yet not fully white and but much of it was extracted using this double extraction method. Hence, a second decoction and extraction were performed using the same method with same cloth but fresh solvent. The process was repeated in the previous manner but this time Sodium Dithionite was used. The first run took around 3 hours and the second run were around 4 hours. The water with dye dispersed into it was taken and collected in another reagent glass bottle and the cloth was kept in another jar.

Dated: **20-02-2025** The collected water with dispersed dye was treated with few drops of 50% Dilute Hydrochloric Acid but no precipitate was found. Hydrochloric Acid was oxidizing the solution rather than acidifying it. Thus, 50% Dilute Sulfuric Acid was added into it until dye precipitate was observed. The alkaline conditions were diminished and dye precipitated out in acidic condition. This mixture was allowed to get filtered through filtration apparatus by Whatman Filter Paper (Grade 42). The precipitate was taken into a glass Petri dish and was dehydrated in oven at 50℃ for 2 minutes. The dehydrated content was then scrapped out and collected as the extracted dye. The weight was approximately: **6g**	Dated: **20-02-2025** The collected water with dispersed dye was treated with few drops of 50% Dilute Hydrochloric Acid but no precipitate was found. Hydrochloric Acid was oxidizing the solution rather than acidifying it. Thus, 50% Dilute Sulfuric Acid was added into it until dye precipitate was observed. The alkaline conditions were diminished and dye precipitated out in acidic condition. This mixture was allowed to get filtered through filtration apparatus by Whatman Filter Paper (Grade 42). The precipitate was taken into a glass Petri dish and was dehydrated in oven at 50℃ for 2 minutes. The dehydrated content was then scrapped out and collected as the extracted dye. The weight was approximately: **10g**

Extracted Dyes into the Solvent Mixture. Collected in different bottles.

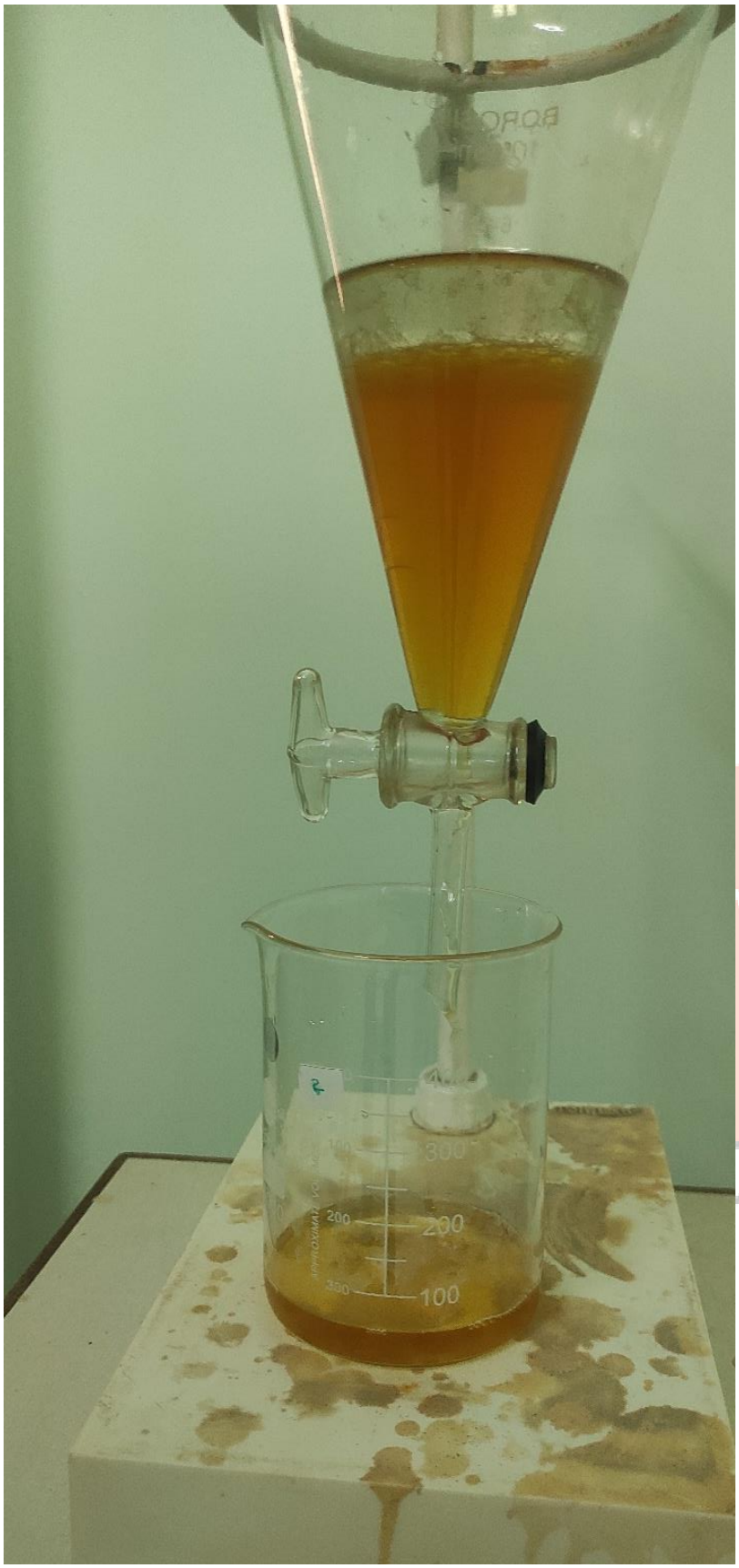




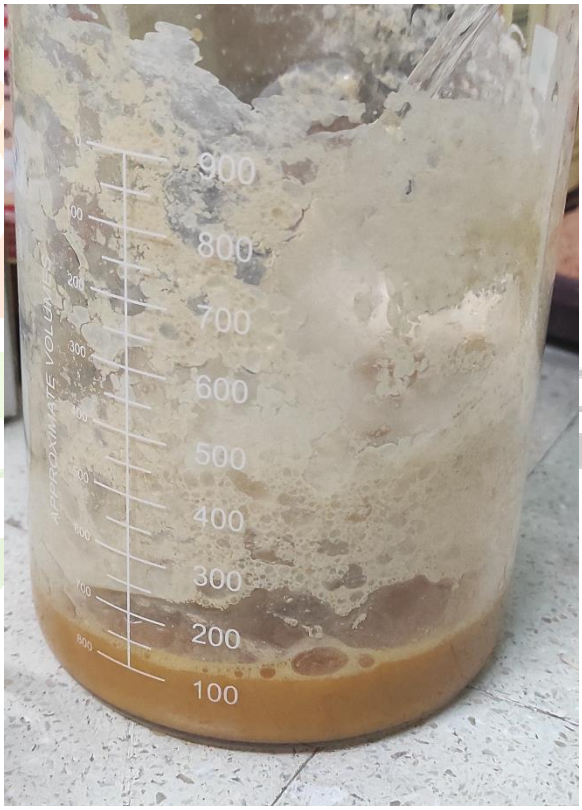
Stirring the organic solvent along with the extracted dye solution; for different dyes.

Seperating the organic solvent from seperating funnel:


300
200



Acidifying Process to get the precipitate:



Acidification causes froth in few solvent mixtures and it settles down to produce a solution with particles precipitated in the bottom. These are further filtered through Whatman Filter paper 42.

Paste Obtained From Filter Paper containing dye.

Note: When we use peroxide and other bleaching agents, the dye is extracted but it is discolored. If bleaching agents are not used; the dye extracted has the same color as the garment.

As the primary results were taken; we could easily sort of the reagents that were able to disperse and extract dyes in a large amount. We could also eliminate those reagents which did not give us any fruitful results. Weak alkaline conditions could not bring us any good until large solvents were added. The reagents which were primarily giving us great results were:

1. Sodium Metabisulfite
2. Sodium Formaldehyde Sulfoxylate
3. Sodium Dithionite

There were solvent conditions which gave us different results. Adding bleaching agents and non-chlorinated bleach could enhance the spontaneity of initial reaction conditions but did not cover up the time limit and were tedious. They might have discolored the fabric initially but the extraction took longer and results were moderately satisfying. These moderately average solvent bases included:

1. Hydrogen Peroxide (*Could Catalyze the reaction; no change on yield*)
2. Sodium Percarbonate (*Discoloration was faster; yield was slow and same*)
3. Sodium Perborate (*Discoloration was faster; yield was slow and same*)
4. Sodium Carbonate (*Acted good with few combinations*)

Thus, another round of testing combinations of these reagents was performed to find out the best suitable match for our extraction process and right conditions for excellent efficiency.

Reagent	**Result**
Sodium Dithionite + Sodium Hydrogen Sulfite	A
Sodium Dithionite + Sodium Metabisulfite	A+
Sodium Dithionite + Sodium Formaldehyde Sulfoxylate	A++
Sodium Metabisulfite + Sodium Hydrogen Sulfite	A

Sodium Metabisulfite + Sodium Formaldehyde Sulfoxylate	A+++
Sodium Hydrogen Sulfite + Sodium Formaldehyde Sulfoxylate	A++

5.3 Identification & Comparison

We have used many reagents for trial basis while performing hot alkaline extraction through the process of Acid-Alkali Extraction. Different alkalis, solvents and reagents were utilized in our research. The table below gives us an idea about what types of reactions took place with which reagents and its proportion and availed us what types of results.

Reagent	Solvent Base	Reaction Mixture	Outcome	Compatibility
Sodium Hydrogen Sulfite ($NaHSO_3$)	Sodium Hydroxide (NaOH)	Distilled water (200ml) + 20g Sodium Hydroxide Flakes (NaOH) + 18g Sodium Hydrogen Sulfite ($NaHSO_3$)	1st Extraction gave about **60-70%** extraction of dye. 2nd Extraction with same solvent and higher reagent gave additional **5%** extraction of dye. 2nd Extraction with new solvent and same amount of reagent gave additional **8-10%** extraction of dye.	**60-70%**
	Sodium Hydroxide (NaOH) + Sodium Carbonate (Na_2CO_3)	Distilled water (200ml) + 20g Sodium Hydroxide Flakes (NaOH) + 18g Sodium Hydrogen Sulfite + 10g Sodium Carbonate (Na_2CO_3)	1st Extraction gave about **70-80%** extraction of dye. 2nd Extraction with the same solvent and higher reagent gave additional **10%** extraction of dye. (*No Sodium Carbonate was added*) 2nd Extraction with new solvent and same amount of reagent gave about **10-12%** additional extraction of dye. 2nd Extraction with new solvent and replacing Sodium hydrogen Sulfite with 10g of Sodium metabisulfite	**70-80%** In Presence of Sodium Metabisulfite: **80-85%**

			($Na_2S_2O_5$) gave about **12-15%** additional extraction of dye.	
	Sodium Hydroxide (NaOH) + Sodium Carbonate (Na_2CO_3) + Hydrogen Peroxide (H_2O_2)	Distilled water (200ml) + 20g Sodium Hydroxide Flakes (NaOH) + 18g Sodium Hydrogen Sulfite + 10g Sodium Carbonate (Na_2CO_3) + 3ml {30%} Hydrogen Peroxide (H_2O_2)	1st Extraction gave about **75-80%** extraction of dye. 2nd Extraction with the same solvent and higher reagent gave additional **12-15%** extraction of dye. (*No Sodium Carbonate was added*) 2nd Extraction with new solvent and same amount of reagent gave about **12-15%** additional extraction of dye. 2nd Extraction with same solvent and same amount of reagent; along with an addition of 10g Sodium metabisulfite ($Na_2S_2O_5$) gave about **15-18%** additional extraction of dye. 2nd Extraction with new solvent and replacing Sodium hydrogen Sulfite with 10g of Sodium metabisulfite ($Na_2S_2O_5$) gave about **18%** additional extraction of dye.	~ **80%** In Presence of Sodium Dithionite & Sodium Hydrogen Sulfite: **90-95%**
	Sodium Hydroxide (NaOH) + Sodium Carbonate (Na_2CO_3) + Sodium Percarbonate ($Na_2CO_3.1.5H_2O_2$) + Sodium Perborate ($NaBO_3.4H_2O$)	Distilled water (200ml) + 10g Sodium Hydroxide Flakes (NaOH) + 10g Sodium Hydrogen Sulfite ($NaHSO_3$) + 5g Sodium Percarbonate ($Na_2CO_3.1.5H_2O_2$) + 5g Sodium Perborate ($NaBO_3.4H_2O$)	1st Extraction gave about **75%** extraction of dye. 2nd Extraction with same solvent and higher reagent gave additional **3-4%** extraction of dye. (*No Sodium Carbonate was added*) 2nd Extraction with new solvent and same amount of reagent	Not More Than: **75%**

			gave additional **10%** extraction of dye. 2nd Extraction with same solvent and replacing Sodium Hydrogen Sulfite with 10g Sodium Dithionite ($Na_2S_2O_4$) gave additional **10-15%** extraction of dye. (*No Sodium Carbonate was added*) 2nd Extraction with new solvent and replacing Sodium hydrogen Sulfite with 10g of Sodium Dithionite ($Na_2S_2O_4$) gave about **15-22%** additional extraction of dye.	In Presence of Sodium Dithionite & Sodium Hydrogen Sulfite: **90%**
Sodium Sulfide (N_2S)	Ammonium Hydroxide (NH_4OH)	Distilled water (500ml) + 10g Sodium Sulfide Flakes (Na_2S) + 6ml {22-25%} Ammonium Hydroxide (NH_4OH)	1st Extraction gave about 6**0-63%** extraction of dye. 2nd Extraction with same solvent and higher reagent gave additional **2-4%** extraction of dye. 2nd Extraction with new solvent and same amount of reagent gave additional **8-10%** extraction of dye. 2nd Extraction with same solvent and replacing Sodium Dithionite with 10g Sodium Formaldehyde Sulfoxylate (CH_3NaO_3S) gave additional **15-18%** extraction of dye.	**60-65%** In Presence of Sodium Formaldehyde Sulfoxylate: **75-78%**
	Sodium Hydroxide (NaOH)	There is no need to perform the reaction of Sodium Sulfide and Sodium Hydroxide as it will react to form Sodium Hydrogen Sulfide. It is better to use a direct reagent of Sodium Hydrogen Sulfite in relative to this reaction. **Thus, The Reaction Is Avoided.**		

Sodium Dithionite ($Na_2S_2O_4$)	Sodium Hydroxide (NaOH) + Sodium Percarbonate ($Na_2CO_3.1.5H_2O_2$) + Sodium Perborate ($NaBO_3.4H_2O$)	Distilled water (200ml) + 10g Sodium Hydroxide Flakes (NaOH) + 10g Sodium Dithionite ($Na_2S_2O_4$) + 5g Sodium Percarbonate ($Na_2CO_3.1.5H_2O_2$) + 5g Sodium Perborate ($NaBO_3.4H_2O$)	1st Extraction gave about **70-75%** extraction of dye. 2nd Extraction with same solvent and higher reagent gave additional **5%** extraction of dye. 2nd Extraction with new solvent and same amount of reagent gave additional **8%** extraction of dye.	**70-78%**
	Sodium Hydroxide (NaOH) + Sodium Carbonate (Na_2CO_3)	Distilled water (320ml) + 20g Sodium Hydroxide Flakes (NaOH) + 18g Sodium Dithionite ($Na_2S_2O_4$) + 10g Sodium Carbonate (Na_2CO_3)	1st Extraction gave about **65-73%** extraction of dye. (*Fast color change was observed but overall yield was not very fruitful*) 2nd Extraction with same solvent and higher reagent gave additional **5-12%** extraction of dye. 2nd Extraction with new solvent and same amount of reagent gave additional **8-10%** extraction of dye. (*Increasing or decreasing the amount of Sodium Carbonate gave no good result*)	**68-70%**
	Sodium Hydroxide (NaOH) + Hydrogen Peroxide (H_2O_2)	Distilled water (300ml) + 15g Sodium Hydroxide Flakes (NaOH) + 10ml {22%} Hydrogen peroxide (H_2O_2) + 10g Sodium Dithionite ($Na_2S_2O_4$)		
	Ammonium Hydroxide (NH_4OH) + Sodium Carbonate (Na_2CO_3)	Distilled water (320ml) + 10ml {22-25%} Ammonium Hydroxide (NH_4OH) + 10g Sodium Carbonate (Na_2CO_3)	1st Extraction gave about **45%** extraction of dye. 2nd Extraction with same solvent and higher reagent gave additional **2-5%** extraction of dye.	**50-55%**

			2nd Extraction with new solvent and same amount of reagent gave additional **8-10%** extraction of dye. (*Increasing or decreasing the amount of Sodium Carbonate gave no good result*)	
	Ammonium Hydroxide (NH_4OH) + Sodium Formaldehyde Sulfoxylate (CH_3NaO_3S)	Distilled water (320ml) + 10g Sodium Formaldehyde Sulfoxylate (CH_3NaO_3S) + 10g Sodium Dithionite ($Na_2S_2O_4$) + 6ml {22-25%} Ammonium Hydroxide (NH_4OH)	1st Extraction gave about **80%** extraction of dye. 2nd Extraction with same solvent and higher reagent gave additional **12%** extraction of dye. 2nd Extraction with new solvent and same amount of reagent gave additional **15-20%** extraction of dye.	**80-90%**
Sodium Formaldehyde Sulfoxylate (CH_3NaO_3S)	Sodium Hydroxide (NaOH)	Distilled water (400ml) + 10g Sodium Hydroxide Flakes (NaOH) + 10g Sodium Formaldehyde Sulfoxylate (CH_3NaO_3S) + 10g Soda Ash	1st Extraction gave about **75-80%** extraction of dye. 2nd Extraction with same solvent and higher reagent gave additional **5-10%** extraction of dye. 2nd Extraction with new solvent and same amount of reagent gave additional **15-20%** extraction of dye.	**80-90%**
	Sodium Hydroxide (NaOH)	Distilled water (400ml) + 10g Sodium Hydroxide Flakes (NaOH) + 10g Sodium Formaldehyde Sulfoxylate (CH_3NaO_3S) + 10g Sodium Metabisulfite ($Na_2S_2O_5$)		
	Ammonium Hydroxide (NH_4OH)	Distilled water (300ml) + 10g Sodium Formaldehyde Sulfoxylate (CH_3NaO_3S) + 6ml {22-	1st Extraction gave about **80-85%** extraction of dye.	**80-90%**

		25%} Ammonium Hydroxide (NH_4OH) + 10g Soda Ash + 10g Sodium Dithionite	2nd Extraction with same solvent and higher reagent gave additional **10%** extraction of dye. 2nd Extraction with new solvent and same amount of reagent gave additional **10-12%** extraction of dye.	
Sodium Metabisulfite ($Na_2S_2O_5$)	Sodium Hydroxide (NaOH)	Distilled water (320ml) + 10g Sodium Hydroxide (NaOH) + 10g Sodium Metabisulfite ($Na_2S_2O_5$)	1st Extraction gave about **70-75%** extraction of dye. 2nd Extraction with same solvent and higher reagent gave additional **5%** extraction of dye. 2nd Extraction with new solvent and same amount of reagent gave additional **8-10%** extraction of dye.	**70-75%**
	Sodium Hydroxide (NaOH) + Sodium Carbonate (Na_2CO_3)	Distilled water (320ml) + 10g Sodium Hydroxide (NaOH) + 10g Sodium Carbonate (Na_2CO_3) + 10g Sodium Metabisulfite ($Na_2S_2O_5$)	1st Extraction gave about **75%** extraction of dye. 2nd Extraction with same solvent and higher reagent gave additional **5%** extraction of dye. 2nd Extraction with new solvent and same amount of reagent gave additional **8-10%** extraction of dye.	**~ 75%**
	Sodium Hydroxide (NaOH) + Hydrogen Peroxide (H_2O_2)	Distilled water (480ml) + 15g Sodium Hydroxide (NaOH) + 10ml {30%} Hydrogen Peroxide (H_2O_2) + 15g Sodium Metabisulfite ($Na_2S_2O_5$)	1st Extraction gave about **70%** extraction of dye. 2nd Extraction with same solvent and higher reagent gave additional **5-10%** extraction of dye. 2nd Extraction with new solvent and same amount of reagent	**70-80%**

			gave additional **15-20%** extraction of dye. 2nd Extraction with new solvent and replacing reagent with Sodium Formaldehyde Sulfoxylate gave additional **15-20%** extraction of dye.	In Presence of Sodium Formaldehyde Sulfoxylate: **85-90%**
	Sodium Hydroxide (NaOH) + Sodium Dithionite ($Na_2S_2O_4$)	Distilled water (320ml) + 10g Sodium Hydroxide (NaOH) + 5g Sodium Dithionite ($Na_2S_2O_4$) + 5g Sodium Metabisulfite ($Na_2S_2O_5$)	1st Extraction gave about **80-85%** extraction of dye. 2nd Extraction with same solvent and higher reagent gave additional **5-7%** extraction of dye. 2nd Extraction with same solvent and higher reagent gave additional **10-15%** extraction of dye.	**80-90%**
Additional Test Performed: **Not Economical but suitable for laboratory purposes with controlled conditions.**				
Sodium Sulfate (Na_2SO_4)	Sodium Hydroxide (NaOH)	Distilled water (300ml) + {10g Benzoyl Peroxide ($C_{14}H_{10}O_4$) dissolved in Acetone C_3H_6O} + 10g Sodium Sulfate (Na_2SO_4)	1st Extraction gave about **90%** extraction of dye. 2nd Extraction with same solvent and higher reagent gave additional **2%** extraction of dye. 2nd Extraction with same solvent and higher reagent gave additional **8%** extraction of dye.	**95%**

A Laboratory controlled extraction technique involving N-N-Dimethylformamide was performed using a reflux condenser. This technique gives us extraordinary result without any reagents added to it. On the other hand, when we add reagents like Sodium Dithionite or Sodium Hydrogen Sulfite; the dispersed dye automatically attaches itself back to the cloth fabric. This is because the N-N- Dimethylformamide is used while fixation of dyes on the fabric. When we give the same condition at reflux action; the dye separates itself automatically without any hard reagent. All you need is an ample amount of solvent of N-N-Dimethylformamide.

For this purpose, we have set an apparatus consisting reflux condenser on the top of a 4 Neck round bottom flask. The head of the flask is of the size 34-35mm with all the other 3 necks having the size of 27-29mm. One of the 3 necks is used with a thermometer holder and another has dropping funnel attached to it. The third neck is sealed with a rubber cork and aluminum foil is wrapped with all the necks as well as head. Since our condenser was having 27-29mm opening; we have attached a converter of 34-35 to 27-29.

The solvent was refluxed for 1 hour straight and the dye was dispersed into the solution very quickly. The reflux action was started at around 100 Deg Celsius as the boiling point of N-N-Dimethylformamide is very high compared to that of water. Thus, the water boils and cools from the reflux condenser whereas the solvent remains in the flask for long time in heated conditions.

The dye was dispersed into the solvent. The solvent was partially removed. When our tested reagents were added to it through dropping funnel; i.e. Sodium Metabisulfite & Sodium Formaldehyde Sulfoxylate; the dispersed dye again attached itself to the fabric.

This proves that N-N-Dimethylformamide is alone able to extract the dye in reflux conditions but when our reagents of hot alkaline extraction techniques are added in same condition; the dye does not disperse but acts opposite and gets attached again to the fabric. Thus, re-polymerization occurs and dye gets bonded to the molecules of fibers through the same reagents which are used as separators in our extraction technique (in presence of N-N-Dimethylformamide).

Using a **4 Neck Round Bottom Flask 2000ml** (*34-35 Head and 27-29 Side Neck*). Thermometer Tube in 1 neck, Cork in the 2nd neck and Dropper in 3rd Neck. A **Reflux Condenser** (27-29) fit on Head of Flask with a **Converter of 27-29 to 34-35** placed in between. **Aluminum Foil** to avoid vaporing.

Set Up for Laboratory Extraction Through Reflux Condenser.

5.4 Consequences

- Sodium Hydrogen Sulfite:
  - ❖ With Strong Alkaline Conditions (Sodium Hydroxide):
    1. 60-70% on basic hot decoction in alkaline condition.

2. 70-75% on introducing Sodium Carbonate alone.
3. 85% on Introduction of Sodium Metabisulfite with Sodium Carbonate
4. 80% on Introduction of Hydrogen Peroxide with Sodium Carbonate
5. 95% on Introduction of Hydrogen Peroxide with Sodium Carbonate & Sodium Metabisulfite. **Replacing Sodium Hydrogen Sulfite with Sodium Metabisulfite increased the yield with 95% in presence of Hydrogen Peroxide.**
6. <75% on Introduction of Sodium Carbonate, Sodium Perborate & Sodium Percarbonate. **Showed bleaching effect; color is discharged but not extracted**)
7. 90% on Introduction of Sodium Carbonate, Sodium Perborate & Sodium Percarbonate along with Sodium Dithionite. **Combination of Sodium Dithionite with Sodium Hydrogen Sulfite increased the yield tremendously**.

Positive Results:

Sodium Hydrogen Sulfite when used as a reagent for extraction of dyes from fabric through hot alkaline extraction process; **works efficiently** in presence of Sodium Dithionite, Sodium Perborate & Sodium Percarbonate.

Sodium Hydrogen Sulfite when used as a reagent for extraction of dyes from fabric through hot alkaline extraction process; **works efficiently** in presence of Hydrogen Peroxide, Sodium Carbonate & Sodium Metabisulfite.

Negative Results:

Sodium Hydrogen Sulfite when used as a reagent for extraction of dyes from fabric through hot alkaline extraction process; **works inefficiently alone** with alkaline base Sodium Hydroxide or Sodium Carbonate.

Sodium Hydrogen Sulfite when used as a reagent for extraction of dyes from fabric through hot alkaline extraction process; **works inefficiently** with non-chlorine bleaches like Sodium Percarbonate & Sodium Perborate.

Inferences:

Sodium Metabisulfite works better than Sodium Hydrogen Sulfite when added in strong alkaline base Sodium Hydroxide for extraction of dyes from fabrics. (in presence of Hydrogen Peroxide & Sodium Carbonate; Glycerin as catalyst)
A Combination of Sodium Dithionite with Sodium Hydrogen Sulfite works tremendously well together for extraction of dyes from fabrics. (in presence of Sodium Perborate, Sodium Percarbonate & Sodium Carbonate)

- Sodium Metabisulfite
  - ❖ With Strong Alkaline Conditions (Sodium Hydroxide):
    1. 70-75% on basic Decoction with hot alkaline condition.
    2. 75-78% on introducing Sodium Carbonate alone.
    3. 70-80% on Introduction of Hydrogen Peroxide
    4. 85-90% on Introduction of Sodium Formaldehyde Sulfoxylate. **Removing Hydrogen Peroxide from reaction and adding Sodium Formaldehyde Sulfoxylate.**
    5. 80-90% on introduction of Sodium Dithionite. **Combining Sodium Metabisulfite with Sodium Dithionite also gave improved results.**
    6. **There was no major increase in yield as such with the addition of Sodium Carbonate**. It might catalyze the process but the overall yield remained the same.

Positive Results:

Sodium Metabisulfite when used as a reagent for extraction of dyes from fabric through hot alkaline process; **works efficiently** better than using Sodium Hydrogen Sulfite alone.

Sodium Metabisulfite when used as a reagent for extraction of dyes from fabric through hot alkaline extraction process; **works efficiently** in presence of Sodium Dithionite and Sodium Hydrogen Sulfite. However, A Mixture of Sodium Dithionite and Sodium Metabisulfite yields best quality extract.

Negative Results:

Addition of Sodium Carbonate gave no additional increase or decrease in the extraction process. It showed bleaching effect but **there was no subsequent increase** in the yield of extracted dye with the addition of Sodium Carbonate with Sodium metabisulfite as reagent.

Inferences:

In Comparison to the combination of Sodium Hydrogen Sulfite and Sodium Dithionite; the combination of Sodium Metabisulfite and Sodium Dithionite yields 10-20% better results.
Thus, A Combination of Sodium Dithionite and Sodium Metabisulfite is Preferred.

- Sodium Dithionite:
  - ❖ With Strong Alkaline Conditions (Sodium Hydroxide):
    1. 65% Average on introduction to Sodium Carbonate
    2. 70-75% on Introduction on introduction of Sodium Percarbonate & Sodium Perborate. **Bleaching effect is seen alongside.**
    3. 75% and nearby on introduction to Hydrogen Peroxide and Sodium Carbonate combined. More Reagent was required and double extraction gave better result with double solvent quantity.
  - ❖ With Weak Alkaline Conditions (Ammonium Hydroxide):
    1. 55% on introduction to Sodium Carbonate. **Very Weak result was seen with weak alkaline condition & Sodium Carbonate did not catalyze the effect of this reaction**.
    2. 85% on introduction to Sodium Formaldehyde Sulfoxylate. Enormous increase in extraction even with small amount of Sodium Formaldehyde Sulfoxylate was seen. **The best proven reagent from all the test**.

Positive Result:

Sodium Dithionite when used as a reagent for extraction of dyes from fabric through hot alkaline extraction process; **works moderately efficient alone** with alkaline base Sodium Hydroxide or Sodium Carbonate.

Sodium Dithionite when used as a reagent for extraction of dyes from fabric through hot alkaline extraction process; **works efficiently** in presence of Sodium Formaldehyde Sulfoxylate and Metabisulfite. However, A Mixture of Sodium Dithionite and Sodium Formaldehyde Sulfoxylate yields best quality extract.

Negative Result:

Sodium Dithionite and a mixture of weak alkaline solvent base Ammonium Hydroxide gave very weak result in presence or absence of Sodium Carbonate. It **worked inefficiently** to extract dyes.

Inference:

Sodium Carbonate showed no tendency to increase or decrease the amount of extraction from the dye under weak or strong alkaline condition.
Thus, Sodium Carbonate does not show any major variation except a byproduct reaction within our extraction process. **It does not contribute on the quantity of dye extracted**.

Sodium Dithionite works **inefficiently in weak alkaline conditions** but works **efficiently in strong alkaline conditions**. It works with Sodium Metabisulfite and Sodium Formaldehyde Sulfoxylate but the best result is seen with a combination of Sodium Formaldehyde Sulfoxylate.

- Sodium Formaldehyde Sulfoxylate:
  - ❖ With Strong Alkaline Conditions (Sodium Hydroxide):
    1. 80% on introduction of sodium carbonate alone. Most efficient among all reagents in extracting dye into the solvent,
    2. 95% on introduction to Sodium Metabisulfite. **This gave us the best combination of all other reagents used in this extraction process**.
    3. 85% on introduction to Sodium Dithionite.
  - ❖ With Weak Alkaline Conditions (Ammonium Hydroxide):
    1. 80-85% on introduction of Sodium Dithionite with Sodium Carbonate. Double solvent amount was to be used and extraction process was slower with large number of released gasses. **Economic conditions were not fulfilled**.

Positive Result:

Sodium Formaldehyde Sulfoxylate when used as a reagent for extraction of dyes from fabric through hot alkaline extraction process; **works excellently efficient alone** with alkaline base Sodium Hydroxide or Sodium Carbonate.

Sodium Formaldehyde Sulfoxylate when used as a reagent for extraction of dyes from fabric through hot alkaline extraction process; **works miraculously** with alkaline base Sodium Metabisulfite or Sodium Dithionite. However, A combination of Sodium Formaldehyde with Sodium Metabisulfite yields best possible result.

Sodium Formaldehyde Sulfoxylate gives positive result in all types of solvent bases but weak alkaline conditions need more solvent in comparison to strong alkaline condition.

Negative Result:

Sodium Formaldehyde Sulfoxylate when used as a reagent for extraction of dyes form the fabrics through hot alkaline extraction process; did not give false results and **NO NEGATIVE RESULTS WERE FOUND**. It works alone and with other combination of reagents very fine.

Inference:

A Combination of Sodium Formaldehyde and Sodium Metabisulfite was concluded to be the best combination for extraction of dye from hot alkaline extraction process. The second runner-up in this test was a combination of Sodium Formaldehyde Sulfoxylate combined with Sodium Dithionite.

The best yield was obtained from the combination of Sodium Formaldehyde Sulfoxylate and Sodium Dithionite.

- Sodium Sulfide:
  - ❖ With Weak Alkaline Conditions (Ammonium Hydroxide):
    1. 63% alone in alkaline solution.

2. 75% on introduction to Sodium Formaldehyde Sulfoxylate. **Since it was the best-known reagent on our above test; it was tested in the presence of weak alkaline solvent base. The effectivity was not up to the mark**.

Positive Result:

Sodium Sulfide and a mixture of our best-known reagent Sodium Formaldehyde Sulfoxylate gave up to 75% of result. The test was positive; yet the yield was less in comparison to Sodium Hydroxide and Sodium Metabisulfite.

Negative Result:

Sodium Sulfide and a mixture of weak alkaline solvent base Ammonium Hydroxide gave average results in presence or absence of Sodium Carbonate. It **worked moderately efficient** in extraction of dyes. However, it consumed a lot of solvent and high emission of toxic gasses was present.

Inference:

Sodium Sulfide worked as a reagent with Ammonium Hydroxide but with a huge quantity of solvent and the effectivity of Sodium Formaldehyde Sulfoxylate was decreased than the previous strong alkaline solvent base condition.

- Sodium Sulfate:
  - ❖ Laboratory Conditions:
    1. 85-95% effectivity on using this laboratory technique; however, it created more complication. The yield was more with expensive reagents and equipment.

Inferences:

This technique is suitable for small quantity laboratory testing but it is tedious and expensive for industrial usage. The time consumed was increased by 3 times.

5.5 Heavy Metal & Toxins Test – Colorimetric Analysis

At the end of our extraction, we found that pink dye from polyester clothing was not able to separate through our process of Hot Alkaline Separation method. Thus, at the end of our journey, we have 6 Samples. These six samples are arrayed in order and tested for heavy metals and toxins present in it. They are numbered in the following manner:

1. Orange Dye – Children Undergarment
2. Green Dye – Children Undergarment
3. Green Dye – Adult Undergarment
4. Black Dye – Female Undergarment
5. Black Dye – Male Undergarment
6. Red Dye – Adult Undergarment

We have performed various test to confirm the presence of different heavy metals or toxins present in the dye. Our observations are as follows:

Dye No.	Toxin	Test Type	Result
1	Lead	Potassium Iodide gives YELLOW Color	**POSITIVE**
	Mercury	Nitric Acid gives BLACK precipitate,	**POSITIVE**

		Ammonia gives WHITE precipitate which dissolves in excess, Potassium Iodide gives REDDISH-BROWN precipitate.	
	Cadmium	Sodium Hydroxide gives WHITE precipitate doesn't dissolve in excess reagent. Schiff's base changes color from ORANGE to YELLOW	
	Chromium	Diphenyl-carbazide in acidic condition give RED-VIOLET color, Lead Nitrate gives YELLOW precipitate Hydrogen Peroxide gives BLUE color which fades Heating with acidified Potassium Dichromate gives RED fumes.	**NEGATIVE**
	Nickel	Dimethylglyoxime gives RED precipitate, Ammonium Sulfide gives GREEN precipitate, Sodium Hydroxide gives PALE GREEN precipitate, Bromine water gives BLACK precipitate.	**NEGATIVE**
	Cobalt	Potassium Nitrate with Acetic Acid gives YELLOW precipitate	**NEGATIVE**
	Zinc	Sodium Hydroxide gives Gelatinous WHITE precipitate and dissolves in excess reagent, Potassium Ferrocyanide gives GRAY-WHITE precipitate, Ammonium Sulfide gives WHITE precipitate, HCL or H_2SO_4 gives POP-SOUND bubbles.	**MILD POSITIVE**
	Bismuth	Potassium Iodide gives YELLOW precipitate, Sodium Hydroxide gives WHITE precipitate	**TRACE POSITIVE**
	Arsenic	Gutzeit Apparatus	**NEGATIVE**

Lead Test

Yellow PPT

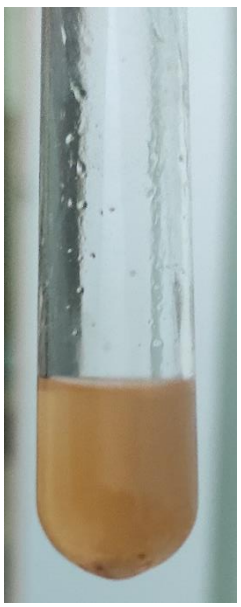
Chromium Test

ABSENT

Nickel Test

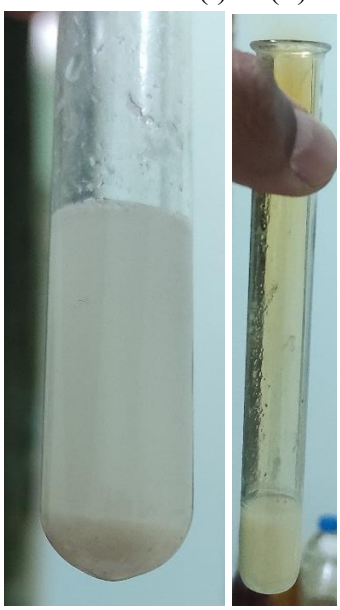
ABSENT

Bismuth Test



Yellow PPT

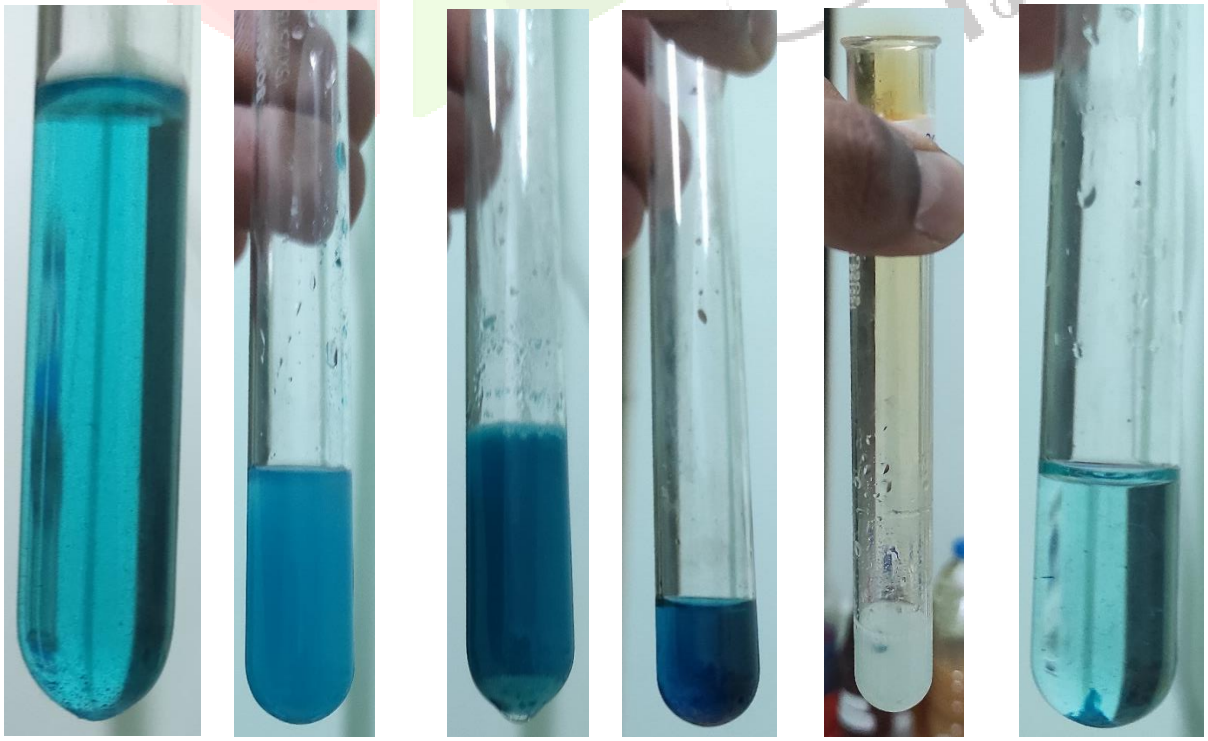
Zinc Tests (i) & (ii)



White PPT & Yellow Fumes

Dye No.	Toxin	Test Type	Result
2	Lead	Potassium Iodide gives YELLOW Color	NEGATIVE
	Mercury	Nitric Acid gives BLACK precipitate, Ammonia gives WHITE precipitate which dissolves in excess, Potassium Iodide gives REDDISH-BROWN precipitate.	POSITIVE
	Cadmium	Sodium Hydroxide gives WHITE precipitate doesn't dissolve in excess reagent. Schiff's base changes color from ORANGE to YELLOW	
	Chromium	Diphenyl-carbazide in acidic condition give RED-VIOLET color, Lead Nitrate gives YELLOW precipitate Hydrogen Peroxide gives BLUE color which fades Heating with acidified Potassium Dichromate gives RED fumes.	NEGATIVE
	Nickel	Dimethylglyoxime gives RED precipitate, Ammonium Sulfide gives GREEN precipitate, Sodium Hydroxide gives PALE GREEN precipitate, Bromine water gives BLACK precipitate.	NEGATIVE
	Cobalt	Potassium Nitrate with Acetic Acid gives YELLOW precipitate	NEGATIVE
	Zinc	Sodium Hydroxide gives Gelatinous WHITE precipitate and dissolves in excess reagent, Potassium Ferrocyanide gives GRAY-WHITE precipitate, Ammonium Sulfide gives WHITE precipitate, HCL or H_2SO_4 gives POP-SOUND bubbles.	MILD POSITIVE
	Bismuth	Potassium Iodide gives YELLOW precipitate, Sodium Hydroxide gives WHITE precipitate	NEGATIVE
	Arsenic	Gutzeit Apparatus	TRACE POSITIVE

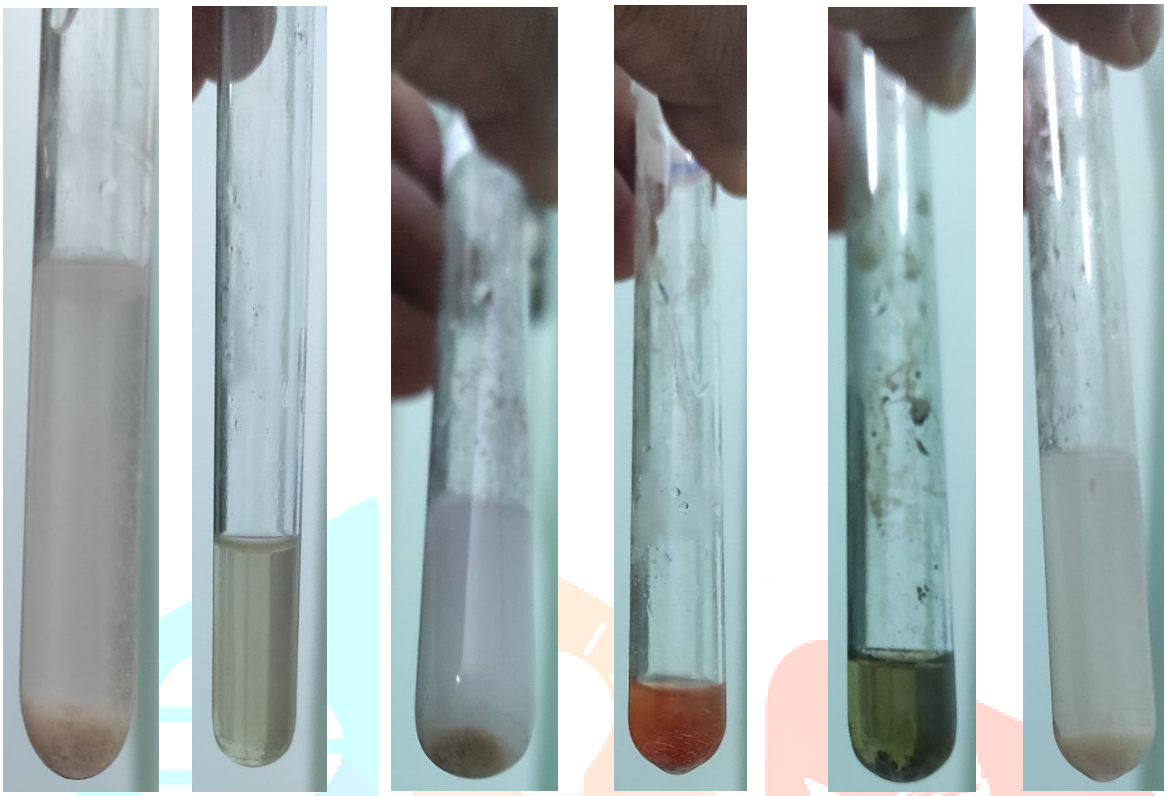
Lead Test Chromium Test Zinc Test (i) Cobalt Test Zinc Test (ii) Bismuth Test



ABSENT ABSENT WHITE PPT ABSENT Yellow Fumes ABSENT

Dye No.	Toxin	Test Type	Result
3	Lead	Potassium Iodide gives YELLOW Color	**POSITIVE**
	Mercury	Nitric Acid gives BLACK precipitate, Ammonia gives WHITE precipitate which dissolves in excess, Potassium Iodide gives REDDISH-BROWN precipitate.	**NEGATIVE**
	Cadmium	Sodium Hydroxide gives WHITE precipitate doesn't dissolve in excess reagent. Schiff's base changes color from ORANGE to YELLOW	
	Chromium	Diphenyl-carbazide in acidic condition give RED-VIOLET color, Lead Nitrate gives YELLOW precipitate Hydrogen Peroxide gives BLUE color which fades Heating with acidified Potassium Dichromate gives RED fumes.	**POSITIVE**
	Nickel	Dimethylglyoxime gives RED precipitate, Ammonium Sulfide gives GREEN precipitate, Sodium Hydroxide gives PALE GREEN precipitate, Bromine water gives BLACK precipitate.	**POSITIVE**
	Cobalt	Potassium Nitrate with Acetic Acid gives YELLOW precipitate	**TRACE POSITIVE**
	Zinc	Sodium Hydroxide gives Gelatinous WHITE precipitate and dissolves in excess reagent, Potassium Ferrocyanide gives GRAY-WHITE precipitate, Ammonium Sulfide gives WHITE precipitate, HCL or H_2SO_4 gives POP-SOUND bubbles.	**POSITIVE**
	Bismuth	Potassium Iodide gives YELLOW precipitate, Sodium Hydroxide gives WHITE precipitate	**MILD POSITIVE**
	Arsenic	Gutzeit Apparatus	**NEGATIVE**

Lead Test Nickel Test Cobalt Test Chromium Test Bismuth Test Zinc Test

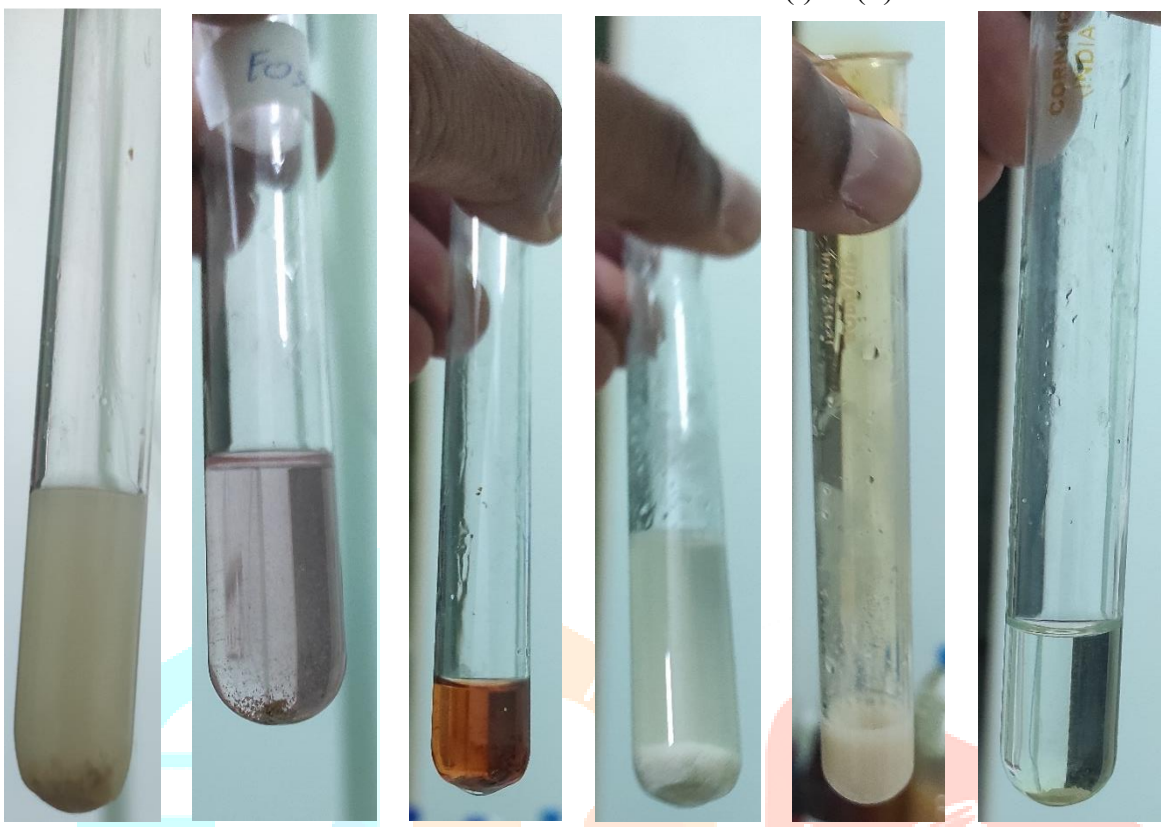


Yellow PPT Pale Green Yellow PPT Red-Violet Yellow Color White PPT

Dye No.	Toxin	Test Type	Result
4	Lead	Potassium Iodide gives YELLOW Color	**POSITIVE**
	Mercury	Nitric Acid gives BLACK precipitate, Ammonia gives WHITE precipitate which dissolves in excess, Potassium Iodide gives REDDISH-BROWN precipitate.	**NEGATIVE**
	Cadmium	Sodium Hydroxide gives WHITE precipitate doesn't dissolve in excess reagent. Schiff's base changes color from ORANGE to YELLOW	
	Chromium	Diphenyl-carbazide in acidic condition give RED-VIOLET color, Lead Nitrate gives YELLOW precipitate Hydrogen Peroxide gives BLUE color which fades Heating with acidified Potassium Dichromate gives RED fumes.	**POSITIVE**
	Nickel	Dimethylglyoxime gives RED precipitate, Ammonium Sulfide gives GREEN precipitate, Sodium Hydroxide gives PALE GREEN precipitate, Bromine water gives BLACK precipitate.	**POSITIVE**
	Cobalt	Potassium Nitrate with Acetic Acid gives YELLOW precipitate	**MILD POSITIVE**
	Zinc	Sodium Hydroxide gives Gelatinous WHITE precipitate and dissolves in excess reagent, Potassium Ferrocyanide gives GRAY-WHITE precipitate, Ammonium Sulfide gives WHITE precipitate, HCL or H_2SO_4 gives POP-SOUND bubbles.	**POSITIVE**
	Bismuth	Potassium Iodide gives YELLOW precipitate,	**NEGATIVE**

		Sodium Hydroxide gives WHITE precipitate	
	Arsenic	Gutzeit Apparatus	NEGATIVE

Lead Test Nickel Test Chromium Test Zinc Test (i) & (ii) Bismuth Test

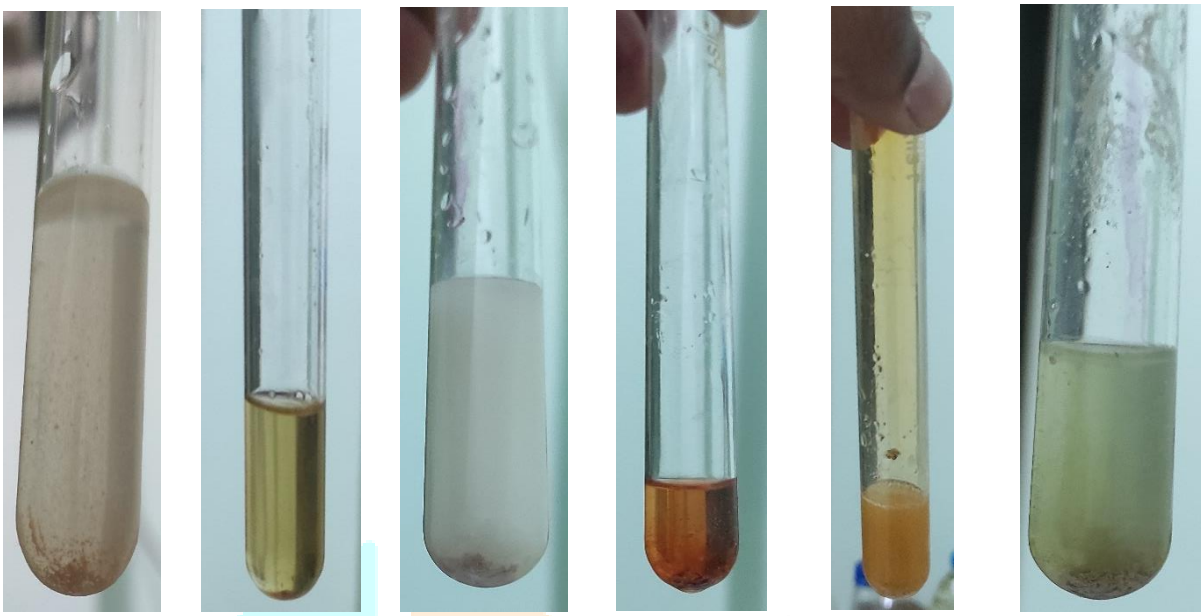


Yellow PPT Red PPT Red-Violet Color White PPT & Yellow Fumes ABSENT

Dye No.	Toxin	Test Type	Result
5	Lead	Potassium Iodide gives YELLOW Color	POSITIVE
	Mercury	Nitric Acid gives BLACK precipitate, Ammonia gives WHITE precipitate which dissolves in excess, Potassium Iodide gives REDDISH-BROWN precipitate.	NEGATIVE
	Cadmium	Sodium Hydroxide gives WHITE precipitate doesn't dissolve in excess reagent. Schiff's base changes color from ORANGE to YELLOW	
	Chromium	Diphenyl-carbazide in acidic condition give RED-VIOLET color, Lead Nitrate gives YELLOW precipitate Hydrogen Peroxide gives BLUE color which fades Heating with acidified Potassium Dichromate gives RED fumes.	POSITIVE
	Nickel	Dimethylglyoxime gives RED precipitate, Ammonium Sulfide gives GREEN precipitate, Sodium Hydroxide gives PALE GREEN precipitate, Bromine water gives BLACK precipitate.	TRACE POSITIVE
	Cobalt	Potassium Nitrate with Acetic Acid gives YELLOW precipitate	HIGHLY POSITIVE
	Zinc	Sodium Hydroxide gives Gelatinous WHITE precipitate and dissolves in excess reagent, Potassium Ferrocyanide gives GRAY-WHITE precipitate, Ammonium Sulfide gives WHITE precipitate, HCL or H_2SO_4 gives POP-SOUND bubbles.	NEGATIVE
	Bismuth	Potassium Iodide gives YELLOW precipitate, Sodium Hydroxide gives WHITE precipitate	HIGHLY POSITIVE

	Arsenic	Gutzeit Apparatus	**TRACE POSITIVE**

Lead Test Zinc Test Nickel Test Chromium Test Cobalt Test Bismuth Test



Yellow PPT ABSENT Red PPT Red-Violet Color Yellow PPT Yellow PPT

Dye No.	**Toxin**	**Test Type**	**Result**
6	Lead	Potassium Iodide gives YELLOW Color	**POSITIVE**
	Mercury	Nitric Acid gives BLACK precipitate, Ammonia gives WHITE precipitate which dissolves in excess, Potassium Iodide gives REDDISH-BROWN precipitate.	**NEGATIVE**
	Cadmium	Sodium Hydroxide gives WHITE precipitate doesn't dissolve in excess reagent. Schiff's base changes color from ORANGE to YELLOW	**NEGATIVE**
	Chromium	Diphenyl-carbazide in acidic condition give RED-VIOLET color, Lead Nitrate gives YELLOW precipitate Hydrogen Peroxide gives BLUE color which fades Heating with acidified Potassium Dichromate gives RED fumes.	**POSITIVE**
	Nickel	Dimethylglyoxime gives RED precipitate, Ammonium Sulfide gives GREEN precipitate, Sodium Hydroxide gives PALE GREEN precipitate, Bromine water gives BLACK precipitate.	**MILD POSITIVE**
	Cobalt	Potassium Nitrate with Acetic Acid gives YELLOW precipitate	**NEGATIVE**
	Zinc	Sodium Hydroxide gives Gelatinous WHITE precipitate and dissolves in excess reagent, Potassium Ferrocyanide gives GRAY-WHITE precipitate, Ammonium Sulfide gives WHITE precipitate, HCL or H_2SO_4 gives POP-SOUND bubbles.	**MILD POSITIVE**
	Bismuth	Potassium Iodide gives YELLOW precipitate, Sodium Hydroxide gives WHITE precipitate	**POSITIVE**
	Arsenic	Gutzeit Apparatus	**POSITIVE**

5.6 Comparative TLC on Sample

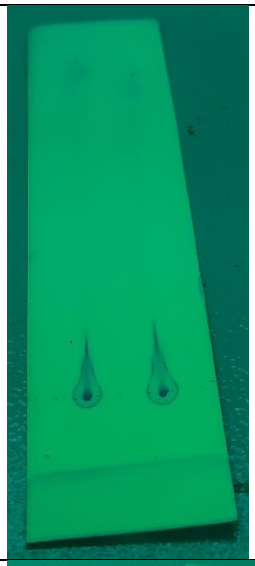
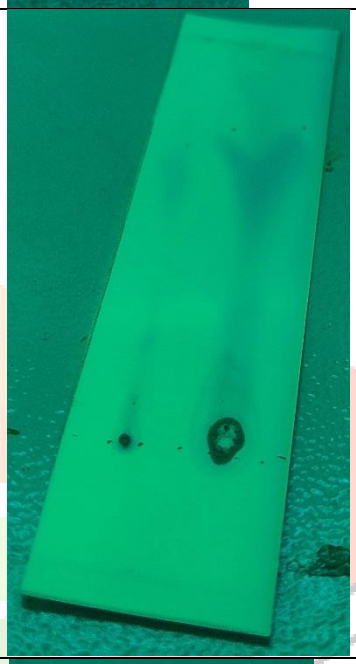
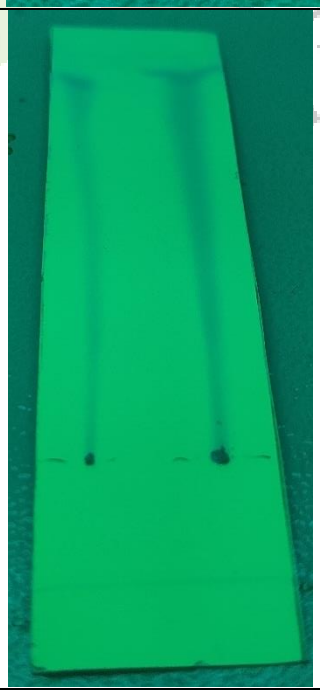
A comparative Thin Layer Chromatography has been performed on these dyes with variable solvent mixture. As stationary phase, Silica Gel coated aluminum thin plate is chosen and as mobile phase various solvents are chosen. These dyes are made to adsorb in the following solvent mixtures differently. The mobile phases chosen are:

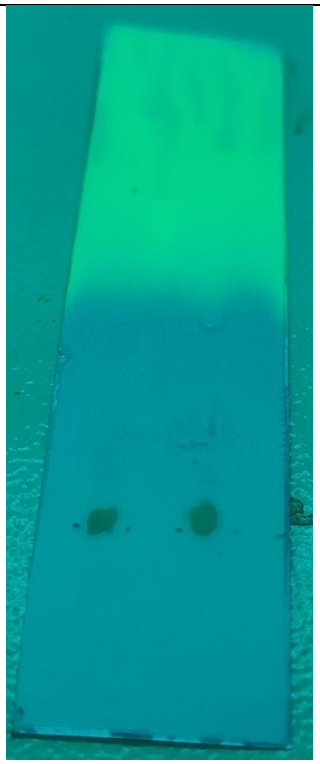
1. Toluene : Pyridine (4:1)
2. 10% Ammonium Hydroxide in Methanol & 10% mixture of this in Ethyl Acetate.
3. Ethyl Acetate : Methanol : Ammonia{0.88} : Water (33:11:5:5)
4. Hexane : Acetic Acid : Water (50:10:20)
5. Hexane : Methanol : Ammonia : Pyridine : Water (8:3:4:4:3)
6. Methanol : Chloroform : Water (5:5:2)

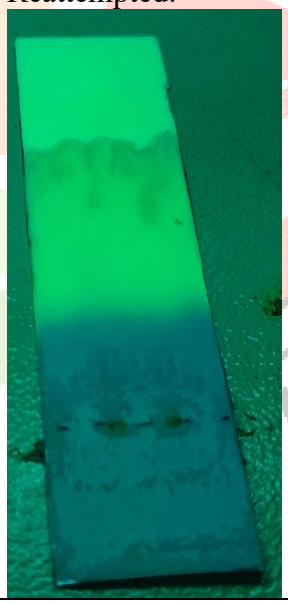
Different dyes were tested on different mobile phases and observations were made. Silica coated aluminum thin layer plates were cut out and markings were made for the spotting for sample of dyes. These dyes were spotted on different TLC plates and different mobile phases were chosen in different beakers. The solvent mixtures were made and they were allowed to rest for a proper mix. After a while, spotting was made with the help of capillary tubes on the lower marking. The marking was made at a distance that it did not dip into the solvent layer in the beaker. It was kept diagonally and it was allowed to get adsorb through the stationary phase. It was kept until the solvent and the solute had risen to the upper marking.

The TLC plates were taken off the solvent beaker and were analyzed under short- and long-range UV light. The distance of travel of solute and solvent was seen and was marked with a sharp pencil. Rf Factor was further calculated and readings were made thereby.

Dye No.	**Solvent Mixture**	**UV**	**Rf Factor**
1	Toluene : Pyridine (4:1)		**For 1st Spot:** Distance Of Solute: 3.5 Distance of Solvent: 7.8 Rh Factor: 0.4487 **For 2nd Spot:** Distance Of Solute: 3.0 Distance of Solvent: 7.8 Rh Factor: 0.3846 Mean Rf: **0.416657**

2	10% Ammonium Hydroxide in Methanol & 10% mixture of this in Ethyl Acetate.		**For 1st Spot:** Distance Of Solute: 6.9 Distance of Solvent: 7.6 Rh Factor: 0.9078 **For 2nd Spot:** Distance Of Solute: 6.4 Distance of Solvent: 7.6 Rh Factor: 0.8421 Mean Rf: **0.874997**
3	Ethyl Acetate : Methanol : Ammonia{0.88} : Water (33:11:5:5)		**For 1st Spot:** Distance Of Solute: 5.0 Distance of Solvent: 7.5 Rh Factor: 0.6666 **For 2nd Spot:** Distance Of Solute: 5.6 Distance of Solvent: 7.5 Rh Factor: 0.7466 Mean Rf: **0.706663**
4	Hexane : Acetic Acid : Water (50:10:20)		**For 1st Spot:** Distance Of Solute: 6.6 Distance of Solvent: 7.3 Rh Factor: 0.9041 **For 2nd Spot:** Distance Of Solute: 6.8 Distance of Solvent: 7.3 Rh Factor: 0.9315 Mean Rf: **0.917803**

5	Hexane : Methanol : Ammonia : Pyridine : Water (8:3:4:4:3)	 Failed Attempt due to Tailing of Chroma. Reattempted: 	**For 1st Spot:** Distance Of Solute: 5.7 Distance of Solvent: 6.6 Rh Factor: 0.8636 **For 2nd Spot:** Distance Of Solute: 5.8 Distance of Solvent: 6.6 Rh Factor: 0.8787 Mean Rf: **0.8712119**

6	Methanol : Chloroform : Water (5:5:2)	 Failed Attempt Reattempted: 	**For 1st Spot:** Distance Of Solute: 3.5 Distance of Solvent: 5.5 Rh Factor: 0.6363 **For 2nd Spot:** Distance Of Solute: 3.4 Distance of Solvent: 5.5 Rh Factor: 0.61818 Mean Rf: **0.627272**

CHAPTER 6: RESULTS

The data showed that most of the basic or acidic dyes were extracted through hot alkaline extraction process from cotton fabrics and semi-cotton nylon fabrics. Polyester fabric dyes are not being eliminated through this process very easily; since they are reactive dyes and react inversely with the solvent and discolor rather than getting extracted. The findings indicate that among all different reagents, a combination of Sodium Metabisulfite and Sodium Formaldehyde Sulfoxylate gave the best yield in double extraction process. Sodium Hydrogen Sulfide is also an equivalent reagent for extraction of fabric dyes but the solvent requirement is increased sufficiently. Double extraction is necessary for ample sample extraction for every dye; may it be acidic or basic.

When we talk about laboratory method, using N-N-dimethylformamide in reflux condition is a very useful extraction technique but it is temperature constraint and requires large solvent and is expensive. Although, it gives us result in single extraction rather than double extraction. Moreover, adding reagents into the same liquid will fix the dye rather than extracting it from the bonds of fiber.

When we talk about hazardous and heavy metals in the dyes; the results imply that Lead, Zinc and Bismuth are present in most of the dyes and some dyes also contain Cobalt, etc. Although one needs to perform instrumental analysis to find quantitative analysis of theses toxic components.

When we performed Thin Layer Chromatography on various samples of dyes with various mobile phases (as applicable on each); we found out that the range of Rh Factor lies between 0.65 to 0.90 on an average. Some selective dyes also have Rh Factor around 0.45.

We found various heavy metals and toxins in these fabric dyes and there is a scope of finding hazardous components in these dyes and hence, one needs to perform further studies on these dyes to find carcinogenicity, quantitative analysis and forbidding various chemicals for day-to-day use in fabric dyes.

CHAPTER 7: FORENSIC SIGNIFICANCE & DISCUSSION

The primary concern which leads us to the conquest of fabric dyes is its safe and secure usage. When it comes to health risks and its effects; we must find out how safe every substance of day-to-day usage lies within our perimeter. Many dyes possess health risk and have been banned due to their carcinogenicity and other effects like irritation, toxicity, etc. However, these have been found in last few years through research that long term exposure to certain fabric dyes may cause health risks and its deteriorating for the skin too. There are also high chances that certain synthetic and chemical agents may react with biological fluids like semen, sweat and vaginal fluid to cause interaction and chemical effects. Thus, this field needs more research to understand the effects of dyes over biological fluids as well as sunlight interaction over it.

Second concern lies with fiber comparison; when we can know that both the fibers are of the same fabric and same color; extraction of dye and its comparative analysis becomes utmost importance to identify and classify it for linking it to the victim, suspect or the crime scene. Hence, during confusion and similarity; extraction of dye and its comparative TLC can give us an idea about the fabric dye / fabric being the same or different. This is another way of confirming the presence of victim or perpetrator on the crime scene.

There are also environmental risks as this fabric dyeing process require a lot of solvent, which are dispersed into water bodies. These solvents are highly toxic and merge with waste water to create a bulk of waste. The water spoilt further travels into agricultural lands and vegetation. This is consumed by people living in the area and the toxicity travels from water bodies to the human bodies. Thus, it is necessary to take care of the environmental damage when we are dealing with such chemical fixators.

Last but not the least, we need to deal with occupational toxicity. The labors who work in manufacturing come in contact with these dyes and heavy metals frequently. This creates long-term chronic effects such as respiratory problems, skin irritation, rashes, carcinogenicity, etc. Thus, it is necessary to take adequate steps to prevent the usage of these toxic dyes in dyeing process and utilize PPE kits and other safety equipment for the same.

However, the prime use remains in characterization of dyes through extraction techniques which shall allow the identification of evidence. Hence, a fast and effective extraction technique as well as analyzing technique is much needed.

CHAPTER 8: CONLCUSIONS

Fiber analysis is a crucial and valid precise method to identify individual characteristic of dyes. Fabric pieces found from arson cases, rape cases or burglary cases can be matched with victim's or perpetrator's fabric through dye analysis. The overall visual analysis and the selection of type of fabric may give is class characteristic for comparison. However, when we talk about extraction of a specific dye from the fabric material and comparing it through methods such as TLC; can give us utmost surety through individual characteristic. The fabric or color may be the same; however, the dye used in each fabric with fixators and its property varies and this becomes our pillar for identifying dyes with surety.

Our research summarizes by providing best reagents for fast and efficient dye extraction through acid-alkali hot extraction process and compares various reagent and its combination. This shall provide us with best suitable reagent and solvent for highest yield and fastest method for extraction of dye from the fabric. This research also helps us to list chemicals and reagents which gave us negative result in extraction process. This research also classifies that which fabric dyes are suitable for acid-alkali hot extraction method and which are those chemicals that shall aid us precisely.

Apart from extraction of dyes, we have also mentioned toxicity analysis through chemical tests; where we tested for heavy metals and toxins. Thus, is gives us a statistical idea about which effluents, toxins and hazardous elements are present in fabric through dyeing process; may it be in minor or trace amounts. Our toxicity analysis is performed by various colorimetric tests and apparatus. This has given us information regarding hazardous substances present in the fabric through dyeing process.

Lastly, we have performed TLC with different mobile phases or solvents to check which solvents might be helpful or comparing the dyes when 2 different dye samples are found in the crime scene. The research gives us an idea about the solvent which are likely to give better results for classifying and categorizing dyes through TLC.

We have thus, extracted the dyes and found the best method as well as reagent used to extract dyes from cotton and semi-cotton fabrics. We have also performed colorimetric analysis to test the presence of toxins and TLC to find suitable mobile phase for a comparative study whenever needed. This research shall aid in identifying, classifying and comparing dyes or fabric found at crime scene.

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