Since the creation of the universe followed by creation of man and civilization, man has come-up with various adaptations, so as to survive in the world. To meet the method of showing the seeds and reaping, the harvest and to save the crops from diseases and pests, man started the search of the compounds to work against such problems. On the basis of trial and error he might had used plants and their various parts as powder or decoctions viz. Neem, chrysanthemum and rotten butter milk to improve the yield.

The pesticide is a material, useful for the mitigation, control or elimination of pests of plants or animals. Pesticides are used principally to reduce parasitism and disease transmission in domestic animals, the loss of crop plants, destruction of processed food, textile and wood products besides parasitism and disease transmission in human. These range frequently stem from the feeding activities of the pests, birds, mice, rabbits, rats, insects, mites, ticks, Eel worms, slugs and snails are recognized as pest (McGraw Hill Encyclopedia, 9th Edn).

Some pesticides are obtained from plants and minerals, a few are by the mass culture of microorganism but most pesticides, however are products which are chemically manufactured. Two outstanding examples of the insecticides are DDT and the herbicide 2,4-D. By the mid 1960’s, it became apparent that the DDT, could be two-edged swords, detailed reviews of the properties, stability, persistence and impact upon all facets of the environment were carried out not only with DDT but with other chlorinated organic
insecticides as well. It is necessary to follow explicitly the directions, restrictions and cautions for use on the label of the product container.

**Classification of Pesticides**

Pesticides are classified into three main categories:

1. **Insecticides**: Chemical compounds used to kill the insect.  
   e.g. BHC, Malathion, etc.

2. **Fungicides**: They are used to control fungal infections.  
   e.g. Copper and Sulpher compounds.

3. **Herbicides**: Chemical which are used to kill certain weeds.  
   e.g. Potassium cyanide, 2,4-D, 2,4-DB, etc.

**Herbicides/Weedicides**

As is true of medical science and other aspects of pest control, the control of weeds by means of herbicides or weedicides has benefitted practically everyone. Millions of people have been relieved of the suffering caused by pollens and poisonous plants.

Use of herbicides was developed chiefly in the twentieth century, than rapidly after 1945 when 2,4-D was introduced. It is a type of growth regulator. The use of chemicals to achieve a measure of control over the environment has become an integral part of technology (Honeycutt, 1994).
Herbicides must be selected and applied with care. It is necessary to follow explicitly the directions, restrictions and cautions for use on the label of the product container (Ware, 1999).

There are certain diseases which can be controlled only by the use of chemical herbicides or weedicides. However, a higher degree of human risks are involved in the use of herbicides (Metcalf and Luckmann, 1994).

Exposure of human being to herbicides may occur in laboratories where these substances are synthesized and examined for their chemical, physical and biological properties. Hence, at this stage proper precautions are generally taken. In chemical plants where herbicides are manufactured and during applications of herbicides to vegetation and crop, opportunity for contact, inhalation and even ingestion of these toxic agents can not be ruled out (Rao and Rao, 2000; Backus, 1992b). In addition to occupational exposure, entry to the food chain of man and animals may occur in the form of residues on raw agricultural commodities like fruits, vegetable and dairy products (Hoar et al., 1986). Exposure may also be through the respiratory tract as a result of contamination of the environment after the spraying operation (Nishioka, 2001). Accidental deaths are reported by herbicide poisoning also (Berwick, 1970; Brahmi et al., 2003). Dermal exposure to herbicide during spraying operation in the fields have resulted in acute poisoning of farm workers.
A great majority of the available herbicides and methods for their use fall naturally into six groups. There are dozens of chemicals, hundreds of formulations and a multitude of methods for killing weeds are available. The following classification of herbicides by method of application has proved extremely useful in discussing weed control problems:

1. **Selective Herbicides**
   - (a) Selective foliage contact sprays
   - (b) Selective foliage translocated sprays.
   - (c) Selective root applications.

2. **Nonselective Herbicides**
   - (a) Nonselective foliage contact sprays
   - (b) Nonselective foliage translocated sprays
   - (c) Nonselective root applications

1. **Selective herbicides** are those, that kill some members of a plant populations with little or no injury to others. Eg. Sulfuric acid, which at proper strength will kill wild mustard in barley or knot weed in young onions.

2. **Non-selective herbicides** are those, that kill all vegetation to which they are applied. Eg. Sodium arsenite solution or aromatic oil. These are used to keep roadsides ditch banks and right-of-way open and weed free (Mcgraw Hill Encyclopaedia, 6 vol.).

These general weed control materials are used to get rid of plants that serve as alternate hosts for insects and plant disease. These herbicides are absorbed by the plant and translocated to
the younger tissues. Human exposure to herbicides is usually estimated by measuring levels in the environment such as air, food, water etc., i.e. environmental monitoring. In some cases information on exposure might be obtained by specific herbicides in the human body, tissues, fluids that is to say biological monitoring or might be done by designing experiments to study herbicide effects on non-target animals and to assess the risk in humans exposed to herbicides (Khogali et al., 2005; Alexander et al., 2007).

Numerous studies have been conducted to determine the mechanism of action of the chlorophenoxy herbicides. The chlorophenoxy herbicides are a group of older herbicides. They are used singly or in combination with other ingredients to control broad leaf weeds in turf-grass, pasture, rights of way, corn, soybean (preplant only), small grains and fence rows. Currently chlorophenoxy herbicides are registered for use. They are 2,4-D, 2,4-DB, MCPA and mecoprop (MCPP).

It is a fact that these toxic herbicides capable of entering the body via the food chain are harmful.

In medical science and of other aspects of pest control, the control of weeds by means of herbicides has benefitted every one. The value of agricultural crops has been increased. Many of the herbicides are used in proprietary mixtures. These are usually marketed to combine the properties of two or more toxicants (McGraw Hill Encyclopaedia, 6th Edition).
2,4-D (Dichlorophenoxyacetic acid)

The concept of modern herbicide technology began to develop in 1900, and accelerated rapidly with the discovery of Dichlorophenoxyacetic acid (2,4-D) as a growth regulator in 1944-1945. Since the introduction of 2,4-D, a wide variety of organic herbicides have been developed and have received wide usage in agriculture, forestry and other industries. Modern usage often combines two or more herbicides to provide the desired weed control. It is an aryloxyalkanoic acid known as a 'phenoxyherbicide'. Once absorbed 2,4-D is translocated within the plant and accumulates at the growing points of roots and shoots where it inhibits growth (The Pesticides Manual, 1994).

There are approximately 100 commercial 2,4-D products registered for use in Florida, packaged singly or in combination with other active ingredients.

Nomenclature:

Structure:

Common Name : 2,4-D (BSI, E-ISO) (m) F-ISO, WSSA);
2,4-PA (JMAF)
Chemical Name: (IUPAC) (2,4-di-cholorophenoxy) acetic acid (I)
(C.A.) I (8 & 9 Cl); Reg. No. [94-75-7]
1,4-D; [5742-19-8]

It is colourless powder. The chemical structure of 2,4-D resembles indole acetic acid, a naturally occurring hormone produced by plant to regulate their own growth. It is a broad leaf herbicide with systemic effect. All forms or derivatives are esters, amine and salts of 2,4-D. These are formulated with solvents, carrier or surfactants and are marked in the form of dust, granules, emulsion, oils, water solutions, gels and water soluble packets in a wide range of concentrations.

Trade Name/Synonyms

Acme LV 4
Agent white
Agricorn D
Bladex - B
Brush Killer - 64
Croprider
Dicofur
Dormon
Fernesta
Fernoxone
Ipaner
Lawn - Keep
Mota Maskros
Moxon
Netagron
Pennamine D
Pielik
Plant guard
Silva Prop 1
Verton 38
Tributan
Weed-B-Gon
Weedaful
Agroxone
Weedar (U.S. EPA, 2008)

**USES**

Its salts and esters are systemic herbicides and used at 0.28-2.3 kg/ha. The highest rate persisting in soil is 30 d. unlike auxins, 2,4-D stays at high level within plants tissues. It is used in wide variety of locations including agricultural, residential and public areas.

The principal use is for the control of broad leaf weeds in cereal crops - including wheat, maize, rice, sorgham, grassland and turf areas (2,4-D Fact Sheet, 1998).

The herbicide is used primarily by cereal crop producers. The forestry industry uses 2,4-D, to suppress the growth of hard-woods and undergrowth in conifer plantations. Another application of 2,4-D occurs along major right of ways (i.e. railway tracks) to control brush. In urban
areas 2,4-D is applied to control broad leaf weeds such as dandelions, ragweed and poison ivy. It is the active ingredient in readily available weed control mixtures, for example Killex.

**How 2,4-D Works**

It is absorbed into a plant through the plant's surface. The weed killer circulates through all parts of the plants and causes abnormal growth blocking the passage for the liquids and nutrients. Subsequently, the roots starve and the plant dies.

2,4-D is readily degraded by microbes in soil and water. Leaching to ground water may occur in coarse-grained sandy soils with low organic content or with very basic soils (Rao *et al.*, 1979).

The area where 2,4-D releases to water and land are - the major industries named:

(i) Cane sugar
(ii) Agricultural chemicals
(iii) Plastics, resins
(iv) Miscellaneous manufacturers
(v) General chemicals

In recent years, a significant increase in the use of 2,4-D against agricultural pests has been observed. One of the major reasons for the increase of using 2,4-D is for ensuring an absolute result. 2,4-D is absorbed by human skin by prolonged contact short-term contact leads to irritation of the skin (Abbott *et al.*, 1987; Pont *et al.*, 2004 and Brand
et al., 2004). It is fact that 2,4-D is capable of entering the body via the food chain and is also harmful to the ecosystem (Ramel, 1977). A way of analysing these illness is to highlight the effects of these harmful chemicals on the enzymes (McComb et al., 1972). Enzymes have a very important role in the metabolic process since they are biological catalysts (Paulino et al., 1991). Their deficiency or surplus indicates various diseases (Rej et al., 1973; Allain, 1974).

**TOXICITY**

(i) Acute toxicity

Acute oral LD$_{50}$ for rats is estimated 375 mg 2,4-D/kg. The toxicity and hazards of the herbicide to man, domestic animals and wild lives are many. 2,4-D is a WHO class II moderately hazardous, pesticides. This places it in the same class as endosulfan, lindane paraquate and toxaphene. Documented health problems are related to 2,4-D include reproductive damage (i.e. sterility) (New York Times, 1970) respiratory difficulties, atrophy, nausea, loss of appetite, skin rashes, eye irritation and chronic headache (Gorzinski et al. 1987). Non-Hodgkins lymphoma has also been associated with 2,4-D exposure (Kogevinas, 1995; Zahm, 1990; Hardwell et al., 1999, 1994; Figg et al., 2000).

Further more, there is evidence of teratogenicity (birth defects) and mutagenecity (mutation of cells) provided by studies involving 2,4-D and lab animals (Environment fact sheet and Nielso et al., 1965).
2,4-D is one of few herbicides to cause nervous system damage (Garcia et al., 2004 and Duffard et al., 1995); both digestion and inhalation affect the central nervous system. Effects to nervous system include inflamed nerve endings, lack of coordination, stiffness in the arms and legs, inability to walk, fatigue, stupor, coma and death (US EPA, 2004; Bortolozzi et al., 1998, 1999a, 1999b; Bortolozzi et al., 2001, 2004).

(ii) Chronic toxicity

It seems that long term exposure to 2,4-D can effect different animals in a wide variety of ways (Kaioumova et al., 2001). It also seems that the various chemical forms of 2,4-D can have different toxic effects. It also causes cellular mutations, which can lead to cancer; phenoxy acid herbicides have been linked with soft tissue sarcomas (Wiklund et al., 1986). The International Agency for Research on Cancer (IARC) has classified 2,4-D among the phenoxy acid herbicides MCPA and 2,4,5-T as a class 2B carcinogen possible carcinogenic to humans (WHO, 1987; Kogenivas et al., 1993; Saracci et al., 1991).

The US authorities have also been reluctant to declare 2,4-D as a potential human carcinogen, but the US courts decided that a forestry workers contracted cancer and died as a direct result of his exposure to 2,4-D during the course of his work (O'Brien, 1987; Woods et al., 1989).

Abnormal foetal skeletal development, increased foetal mortality and other reproductive effects are fairly conclusively associated with
exposure to phenoxy acid herbicide and their dioxin contaminants (IPCS, Geneva, 1984; Arbuckle et al., 1998 and Arias, 1994). 2,4-D has also classified as endocrine disrupter (EHP, 1993 and Haddow et al., 1999). A mounting body of evidence links 2,4-D to various cancers (Reuber, 1983 and An SAB Report, 1994), particularly Non-Hodgkin's lymphoma (Ballester 1993). EPA has been reluctant to classify it as a carcinogen in the face of industry pressure.

The risk of cancer was higher for farmers who mixed or applied the pesticide themselves (Hallenbeck et al., 1985; Saracci et al., 1991; Lynge, 1985). The herbicides can cause rare brain tumors in rats.

Long term exposure to 2,4-D also results in a wide range of other health problems. Chronic oral intake results in lesions of the kidney and liver in both rats and dogs (Cox, 1999).

In human, two studies showed a connection between hepatitis cases and chronic oral consumption of 2,4-D by golfers, who habitually licked their golf balls (Johnston et al., 1998).

2,4-D is also an endocrine disrupter (Colbon, 1993), a chemical that can interfere with the body's hormone messaging system and can alter many essential processes. It is also a suspected endocrine disruptor (NIHS, 2004).

In studies with rats 2,4-D has been shown to alter levels of metabolism and sex hormone (Liu et al., 1996). Reproductive toxicity has also been observed in relation to 2,4-D. Residues of 2,4-D are
detectable in urine and semen samples of men who apply the herbicide (Health Canada, 1999). In rats, exposure resulted in tissues with abnormal cavities, bleeding, increased mortality and genetic damage (ETN, 1996).

It also found that the birth defect rate to be highest in areas where 2,4-D used highest. Another study conducted in 2003 examined the wheat producing states where more than 85% of the acre-age is treated with chlorophenoxy herbicides, including 2,4-D. Children conceived during the time of herbicide production were more likely to have birth defects (Schreinemachers, 2003).

<table>
<thead>
<tr>
<th>Common Name</th>
<th>Rat oral LD$_{50}$</th>
<th>Rabbit dermal LD$_{50}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>2,4-D</td>
<td>500-949</td>
<td>-</td>
</tr>
<tr>
<td>2,4-DB</td>
<td>&gt;2,000</td>
<td>&gt;10,000</td>
</tr>
</tbody>
</table>

Chlorophenoxy herbicide (mammalian toxicities: mg/kg of body weights)

**Environmental Effects**

2,4-D has been shown to have negative impacts on a number of animals. The toxicity of 2,4-D to fish is variable. The butoxy-ethanol ester is very toxic to fish, but other forms are less toxic. 2,4-D also bioaccumulates in fish, meaning that fish tissues will contain a higher concentration of 2,4-D than the surrounding water and puts them at even greater risk. Consumption of plants treated with 2,4-D killed cattle and horses (Cox, 1999).
Chapter-1: Introduction

2,4-D has been shown to cause cellular mutations, which can lead to cancer. This mutagen contains dioxins, a group of chemicals known to be hazardous to human health and to the environment (Hayes et al., 1991).

2,4-D is a moderately persistent chemical with a half life between 20 and 200 days. Unfortunately, the herbicide does not affect target weed alone. It can cause low growth rates, reproductive problems (Grover et al., 1991) changes in appearance or behaviour or death in non-target species. Additionally the spraying of 2,4-D often, contaminates water system, about 91.7% of 2,4-D will eventually end up in water. This contamination threatens the vegetation and the animal life that consumes it. The chemical will also be carried by run-off into the local river-system thereby jeopardizing the health of aquatic life as well (Sierra club of Canada, 2005; Alexander et al., 1985). In the urban setting, it has been proven that house holds using 2,4-D put their dogs at twice the risk of developing canine malignant lymphoma (Hayes et al., 1995).

Regulatory Status and History

2,4-D was one of the first herbicides to be commercially marketed. It was first introduced in the United States in the late 1940's. 2,4-D made-up a major portion (about 50%) of the herbicide known as agent orange, which was used during the Vietnam war. However, it is thought that one of the other major component 2,4,5-T was the main
culprit and has now been banned, several forms of dioxin have also been found in 2,4-D, including 2,4,7,8-TCDD.

The history of dioxin contamination further increases the dangers related to 2,4-D, particularly for the amine and ester forms dioxin’s are highly carcinogenic and can cause health problems as severe as weakening of the immune system, decreased fertility, altered sex hormones, miscarriage, birth defects and cancer. U.S. EPA studies in 1994 detected dioxins in a number of 2,4-D products. The Washington Department of Agriculture (1998) also detected dioxin in a 2,4-D product.

2,4-D is currently undergoing EPA’s reregistration process. According to USEPA the Registration Eligibility Decision (RED, 2005) on June 23, 2004, the EPA released the Public Series of risk assessment documents summarizing current data on the human health and environmental effects of 2,4-D.

With the initial development of pesticides, their use was expansive. The number of scientific studies are showing that the agricultural usage of pesticides have negative ecological effects and toxic effects on living things in the environment and on the immune system (Lee et al., 2001) and different tissues (Sharma et al., 2005; Kalipci et al.; 2010c). Only in the past two decades it has been openly acknowledged that these chemicals are poisonous to humans and their environment.
Since 1980, Agriculture and Agri-Food Canada have been reviewing the regulatory status of 2,4-D. 2,4-D is a herbicide that has been heavily used in agriculture all over the world.

**2,4 DB or BUTOXONE**

2,4-DB was first registered in the U.S. in 1958, its chemical name if butyric acid. It is registered for use as a foliar treatment to peanut and soybean. It is also formulated as amines and low-volatile esters. 2,4-DB is not highly phytotoxic, per se; however, it undergoes beta-oxidation within plants and soils to form 2,4-D. Some plants are able to make this conversion rapidly, resulting in injury or control, while others, such as peanut and soybean, react slowly making them more tolerant of the effects.

2,4-DB is a selective systemic herbicide in the phenoxy family. This compound is not to be confused with another phenoxy compound 2,4-D and its derivatives or with the derivatives of 2,4-DB such as the sodium salt, the isooctyl ester or the butyl ester. Each of these are slightly different compounds and thus would have different toxicities and environmental characteristics.

**Nomenclature**

**Structure:**

\[
\text{Cl}\supseteq\text{O(CH}_2\text{)}_3\text{CO.OH} \text{Cl}
\]
**Common Name:** 2,4-DB (BSI, draft E-ISO, (m) draft F-ISO, WSSA)

**Chemical Name:** (IUPAC) 4-(2,4-dichlorophenoxy) butyric acid (I);
(C.A.) 4-(2,4-dichlorophenoxy) butanoic acid (9CI);
(I) (8CI); Reg No. [94-82-6] 2,4-DB; [10433-59-7]
2,4, DB - Sodium; [19480-40-1] 2,4-DB-Potassium;
[2758-42-1]

**Trade name and Synonyms**
Alistell
Butoxone
Butoxone -200
Butoxone Ester
Butyrac
Clovacorn Extra (FCC)
Clovacorn Plus
Embutox E
Embutox Plus
Nintex
Perselect
Res cue
Venceweed

2,4-DB is 4(2,4-Dichlorophenoxy) butyric acid. It may also be found in formulations with other herbicides such as cyanazine, MCPA, benzolin, linuron and wecoprop.
USES

2,4-DB is manufactured as an acid (2,4-DB) and an amine salt (2,4-DB-DMAS). It is a translocatable herbicide effective against broadleaf weeds but is more selective than 2,4-D because its activity is dependent on beta-oxidation within the plants. It is a systemic herbicide used to control alfalfa, clover peanuts, soyabean, peppermint, spearmint and birds foot trefoil. Approximately 375,000 pounds of 2,4-DB and 2,4-DB-DMAS is used annually. It may be used on lucerne, undersown cereals and grassland at 1.5-3.0 kg 1.l./ha alone or in mixture with MCPA. There are six products containing 2,4-DB (Four technical products and two end use products) and 15 products containing 2,4-DB-DMAS (one formulation intermediate and fourteen end-use-products). There is one section 24C special local need registration for 2,4-DB-DMAS use on mint in Idalio. All end-use product formulations are liquid (EPA, 2005).

How 2,4-DB Works

In soil 2,4-DB is broken down by the action of soil microbes to the product 2,4-D. The half-life for 2,4-DB is about 7 days. The compound is taken up by the roots and moved throughout the plant. In plants, the compound 2,4-DB is degraded to 2,4-D, which is then broken down further to less toxic material. In plants tolerant to the herbicide, the breakdown from 2,4-DB to 2,4-D is very slow (Kidd and James, 1991).
Toxicity

Acute Toxicity

2,4-DB is placed in the following acute toxicity categories. Predicted residues from all uses of 2,4-DB are below the acute level of concern for mammals (Bradbery et al., 2000 and ETN, 1996).

Slight liver hypertrophy was seen in rats (Kidd and James, 1991). Repeated over exposure may cause effect to kidneys, liver, heart, thyroid, adrenal, blood chemistry, stomach and reduced body weight.

Chronic Toxicity

Decrease in body weight, heart weight, changes in blood chemistry, lower ovarian weight and fewer offspring born. The highest dose offspring mortalities and growth reductions were observed during lactation. Embryotoxic effects were also observed with this herbicide (Roll et al., 1983). The incidence of brown pigment in the kidneys was increased in both sexes. Indicative of slight degeneration of the tubular epithelium and renal toxicity were also recorded (IET, 1984).

Studies in laboratory animals with 2,4-DB have shown early resorptions decreased fetal body weights and skeletal variations in the offspring at doses toxic to mother animals. These are considered to result from maternal toxicity. Chromosome changes occurred due to exposure to 2,4-DB in Chinese hamster cells.
Environmental Effect

Available data indicates that ecological risks were only assessed for 2,4-DB (US EPA, 1988). In soil environments 2,4-DB dissipation is dependent on leaching and on oxidative microbial - mediated degradation. The primary route of dissipation is transformation with the major transformation product being 2,4-D. 2,4-DB is not expected to bioaccumulate in the environment.