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Review

Systemic Analysis of Glyphosate Impact on Environment and Human Health

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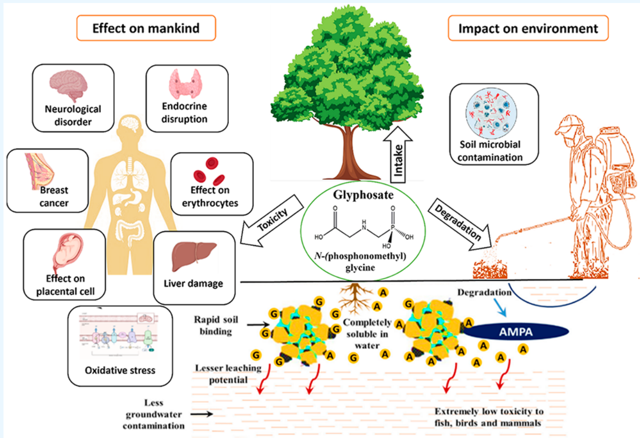
ABSTRACT: With a growing global population, agricultural scientists are focusing on crop production management and the creation of new strategies for a higher agricultural output. However, the growth of undesirable plants besides the primary crop poses a significant challenge in agriculture, necessitating the massive application of herbicides to eradicate this problem. Several synthetic herbicides are widely utilized, with glyphosate emerging as a potential molecule for solving this emerging issue; however, it has several environmental and health consequences. Several weed species have evolved resistance to this herbicide, therefore lowering agricultural yield. The persistence of glyphosate residue in the environment, such as in water and soil systems, is due to the misuse of glyphosate in agricultural regions, which causes its percolation into groundwater via the vertical soil profile. As a result, it endangers many nontarget organisms existing in the natural environment, which comprises both soil and water. The current Review aims to provide a systemic analysis of glyphosate, its various effects on the environment, its subsequent impact on human health and animals, which will lead us toward a better understanding of the issues about herbicide usage and aid in managing it wisely, as in the near the future glyphosate market is aiming for a positive forecast until 2035.



1. INTRODUCTION

Over the past decade, the usage of synthetic chemicals in the agricultural sector has proven to have a catastrophic impact on the environment, leading to the accumulation of harmful residues in the biosphere. The imprudent utilization of pesticides in agricultural fields has led to a paradigm shift in the environment. Though the shear exploitation of agrochemicals has led to an enhancement in agricultural produce by effectively managing the pests, rodents, and microbial infections, such haphazard usage of synthetic chemicals and excessive spraying of these chemicals in the fields has adverse impacts not only on the environment but also to human health and other nontargeted organisms. The modern cultivation practices exploit various synthetic chemicals such as herbicides, insecticides, and pesticides to increase agricultural efficiency. The advent of "agricultural evolution" and the use of pesticides and herbicides was mainly started for mankind, but growing human demands and avidity to increase crop yields has gradually led to their overuse.[1] The discovery of *N*-(phosphonomethyl)glycine was among the dynamic revolutions in agriculture, making it one of the extensively employed herbicides all over the globe. It is typically administered to plants in the form of complicated formulations that promote absorption,[2] with an approximate worldwide demand of half a million tons per annum.[3] This broad-spectrum synthetic organophosphonate herbicide is extensively utilized to destroy undesired plants in agriculture as well as the nonagriculture sector.[4]

For over a decade, glyphosate has been considered as the most effective farming tool and safest herbicide employed in the agriculture industry. In the beginning, glyphosate was observed to exhibit low toxicity to human beings, as there were barely indications of genotoxicity or carcinogenicity in mammals.[5] In 1950, Henri Martin of Swiss pharmaceutical company was among the first one to synthesize *N*-(phosphonomethyl)glycine,[6] and its herbicidal activity on perennial weeds was further tested and confirmed by Monsanto in 1970 and Baird in 1971, respectively.[7] Due to the competition among other herbicides in the market, glyphosate use in agricultural applications was modest in the

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late 1980s and early 1990s. Herbicides such as glyphosate and its derivatives have gained popularity in the United States since 1974, with worldwide usage increasing up to 15-fold owing to the widespread perception that they exhibit no detrimental effects on animals.[4]

However, the persistence of glyphosate and AMPA residues after degradation leads to the deterioration of environmental and soil conditions,[8] as depicted in the Figure 1. Moreover,

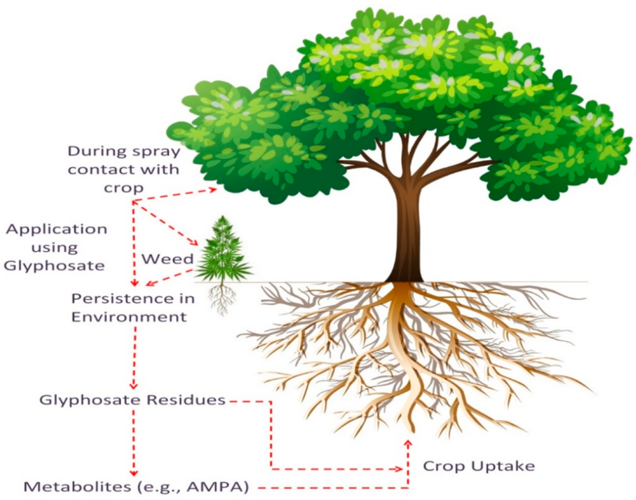

Figure 1. Persistence of glyphosate residues in the environment (e.g., AMPA) is due to its excessive usage in agricultural fields to eradicate weeds.

application of glyphosate and glyphosate-based herbicides (GBHs) has led to the introduction of approximately 38 glyphosate-resistant (GR) weeds.[9] The half-lives of glyphosate and aminomethylphosphonic acid (AMPA) range from 0.8 to 151 days and from 10 to 98 days, respectively.[10] Major worries have been growing regarding the adverse impacts of glyphosate and AMPA on humans, plants, and animal health[11] due to the heavy usage and accumulation of herbicides in environmental reservoir and food sources.[12] Additionally, glyphosate residues in effluents are too difficult to remove, as they persist for a long time in water and soil.[13] *Escherichia coli*, *Salmonella spp.*, and soil bacteria in general have been demonstrated to develop antibiotic resistance when exposed to glyphosate and AMPA.[14] There is numerous evidence of toxicity to fish, birds, amphibians, bees, and other animals.[15−18] Furthermore, investigations indicate that exposure to GBHs, even at low doses, can be teratogenic, carcinogenic, tumorigenic, and hepatotoxic, therefore proving that it leads to oxidative stress.[19]

Furthermore, substantial data suggests that glyphosate may have a negative impact on health, necessitating further monitoring.[20] As a result, it is critical to investigate the possible risks associated with glyphosate, accept the relevance of public issues, and properly balance the concern alongside agricultural benefits.[21] Nevertheless, the constant large consumption and excessive usage over the last 40 years have raised alarm, highlighting its consequences on the environment, human health, and other nontargeted species and emerging as a major concern in the agricultural industry throughout the world. Glyphosate's significance is currently jeopardized due to the emergence of glyphosate-resistant weed species and toxic algae.

2. CHEMICAL STRUCTURE AND MECHANISM

2.1. Chemical Properties. Glyphosates contain a simple yet strong chemical structure pertaining to the organophosphorus compound family.[22] Glyphosate is a phosphonic acid, which is usually formed by oxidative coupling of the methyl group of methyl phosphonic acid with the amino group of glycine.[23] It is well-known that methyl phosphonic acid, which is one of the components of glyphosate, possess qualities that are both corrosive and irritating in nature.[24] Methyl phosphonic acid in its derivative form of dimethyl hydrogen phosphate is known to cause lung lesions, carcinoma, and neoplastic and non-neoplastic lesions in the fore stomachs of male rats.[25] Glyphosate toxicity is also attributed with its various formulations consisting of salts, surfactants, and numerous other minor components. Therefore, toxicity in humans is not due to the active ingredient alone but with complex mixtures. Some of the most commonly used salts are aqueous mixtures of isopropyl amine (IPA) salts of glyphosate and a surfactant such as polyethylene amine (POEA), which increases its hydrophilicity, as they help in the absorption of the herbicide into the leaf and other vegetative parts.[26] The glyphosate structure is distinct among various herbicides due to its straight carbon chain that has a weak bond as compared to various herbicides that consist of an aromatic cyclic structure.[23] With respect to the chemical structure as shown in Figure 2, glyphosate typically behaves as a zwitterion[23] that

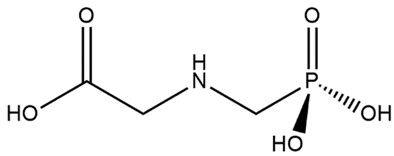

Figure 2. Chemical structure of glyphosate.

possess different functions such as carboxylate, phosphonate, and amine. This zwitterionic nature of glyphosate allows it to bind with metals.[27−29] It does not possess any chemical group in its structure, therefore barring it from binding with deoxyribonucleic acid (DNA) and thereby reducing the risk of chromosomal damage or mutagenicity.[30]

It is a relatively stable with respect to photodecomposition and chemical decomposition.[31] On heating it leads to the production of nitrogen oxides and phosphorus oxides,[32] whereas in soil and water it generally degrades into AMPA and glyoxylate or is converted to glycine, which is a rare route.[33,34] The low vapor pressure of glyphosate (5.7×10^{-8} Pa at 25 °C) has been noted, suggesting that soil volatilization is not a significant form of dissipation.[52] The distinct chemical properties of glyphosate are discussed below in Table 1.

2.2. X-ray Crystallography Structure of Glyphosate. The monoclinic structure with a $P21/c$ space group, according to X-ray crystallography, was observed in glyphosate. The following are the cell diameters for the crystal: a = 8.682(5) Å, b = 7.973(8) Å, c = 9.875(5) Å, β = 105.74(4)°, V = 675.9(8) $Å^3$ and Z = 4. In glyphosate, the various bond lengths (Å) are recorded as follows: the length of the C−P bond is 1.823 Å, the lengths of the C−H and N−H bonds range from 0.914 to 1.118 Å, and the lengths of the O−H bonds in the phosphono ($-P(=O)OH_2$) and carboxyl groups (COOH) are 0.711 and 0.743 Å, respectively. The bond lengths of two O atoms of the phosphono group in the glyphosate molecule, P−O1 = 1.500 Å

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Table 1. Glyphosate's Chemical Properties

s. no.	description	properties
1.	IUPAC name	*N*-(phosphonomethyl) glycine
2.	formula	$C_3H_8NO_5P$
3.	molecular mass	169 g/mol
4.	stability	32 days at 25 °C; pH 5, 7, and 9
5.	density	1.74 g/mL
6.	physical state	32 days at 25 °C; pH 5, 7, and 9, white crystalline solid
7.	melting point	189.5 °C
8.	solubility	soluble in water
9.	amphoteric nature	complex, polyprotic molecule
10.	dissociation constant	four ionizable functional groups: pKa1 = 2.0, pKa2 = 2.29, pKa3= 5.96, and pKa4 = 10.98
11.	odor	odorless

and P−O2 = 1.501 Å, suggest that they are in resonance with one another. The third P−O bond, on the other hand, is a single bond with a bond length of 1.576 Å. Various glyphosate molecules are connected to one another via H-bonds.[35]

2.3. Worldwide Consumption and Global Glyphosate Market Forecast until 2035. Glyphosate acts as a broad-spectrum, nonselective, and post-emergence herbicide utilized to eradicate weeds, mainly broadleaf weeds and grasses that grow with crops and exhibit low mammalian toxicity.[12] The United States (USA) is the largest growing consumer of glyphosate,[36] as farmers used nearly 6−8 million pounds of glyphosate in 1987.[37] Furthermore, 20 years down the line, studies conducted by the Environmental Protection Agency (EPA) in 2007 revealed that glyphosate usage has grown drastically to 81.6−83.9 million kilograms.[38] In 2018, around 35−39% of the agriculture sector is dependent on glyphosate usage in Denmark and Germany.[39] In Argentina, the annual usage of glyphosate as an herbicide is 180−200 million liters.[40] Glyphosate is a common herbicide used throughout agricultural food crops and commercial fruit orchards in the United Kingdom. Glyphosate is used to treat 50−60% of sunflower crops in European nations such as France, Romania, and Hungary each year.[33]

The field of agriculture plays a significant role in the Indian economy. As a direct consequence of the fact that agriculture is the primary source of income for more than 70% of the country's population, glyphosate is utilized on a consistent basis in India. According to a survey, the amount of glyphosate that was used annually in India in 2015 was 35 kilotonnes, and it is anticipated that this number will rise to 75 kilotonnes by the year 2024.[41] The development of crops that are resistant to glyphosate has resulted in increased application of this herbicide all over the world by many fold.[4] Annual glyphosate consumption had reached to 240 million pounds by 2014, which has been concluded from various survey conducted.[16]

The Globe Newswire report "Glyphosate Market-Forecasts from 2022 to 2027" states that the glyphosate market was worth USD 9.016 billion in 2020 and is predicted to increase at a compound annual growth rate (CAGR) of 5.10% during the forecast time frame reach up to USD 12.771 billion by 2027 at CAGR of 6.9%.[42] According to a business research company report, the glyphosate market is expected to increase at a CAGR of 6.32% from 2016 to 2022. Due to coronavirus disease 2019 (COVID-19), glyphosate market has reached only up to $9.02 billion in 2022 as the agricultural sector was moderately functional, further increased to $9.67 billion by 2023 at CAGR of 7.2%, and is expected to grow up to $12.65 billion in 2027.[43] According to a Zion Market research report, the global glyphosate market will be expected to reach up to $10.88 billion in 2024 with a CAGR of 6.2% between 2014−2018.[44] Further, the global glyphosate usage is estimated to $18 billion in 2035, which is supposed to be growing with a CAGR of 6% over the forecast period of 2023−2035.[45] The forecast of glyphosate market analysis on the basis of supply and demand highlights until 2035 is illustrated in Figure 3.

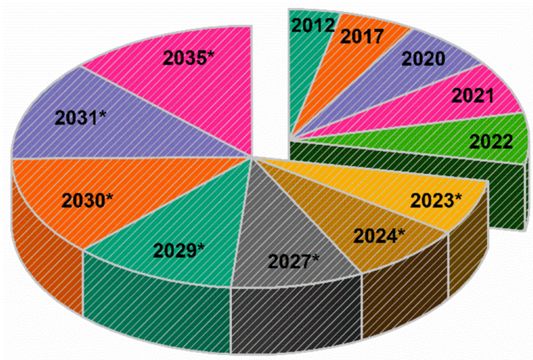

Figure 3. Global glyphosate market analysis forecast until 2035 (*projected glyphosate market analysis according to various reports published).

It has been observed that since 1959 the worldwide population has more than doubled and increased from 3 billion to 6 billion people. As a result, the present rate of global population growth will increase by approximately by 1.05%. Each year, the population will grow by 81 million people, on average. The population is expected to surpass 9 billion by 2037, increasing by 50% during the following 40 years.[46] The current trend reflects the rising worldwide demand for food items. According to the Food and Agriculture Organization's (FAO's) worldwide prospective analysis, food consumption is expected to rise by 59−98% by 2050.[47] To fulfill this need, herbicides like glyphosate are used to eliminate weeds that are harmful to developing crops and plants, boosting demand. The commercialization of genetically engineered−herbicide tolerant (GE-HT) crops and increased glyphosate application in both agricultural and nonagricultural sectors are the major drivers of expansion.[46] According to the U.S. Department of Agriculture (USDA), GE-HT crops currently account for around 56% of worldwide glyphosate consumption, with enhancement in the global market of glyphosate with respect to demand and supply analysis.

The global glyphosate usage by different countries accounts for the fact that its marketplace throughout the United States was expected to be worth around $2.4 billion by 2022. China, the second-largest economy in the world, will reach a market value of $2.2 billion by 2030, rising at a CAGR of 5.7% during 2022 to 2030. Other significant geographic markets comprise Japan and Canada, which are predicted to grow at 3.2% and 3.9%, respectively, from 2022 and 2030. Within Europe, the United Kingdom is expected to develop at 2.8% CAGR. The market in the Asia-Pacific region is expected to generate $1.5 billion by 2030, with nations such as India, Australia, and South Korea leading the way.[48]

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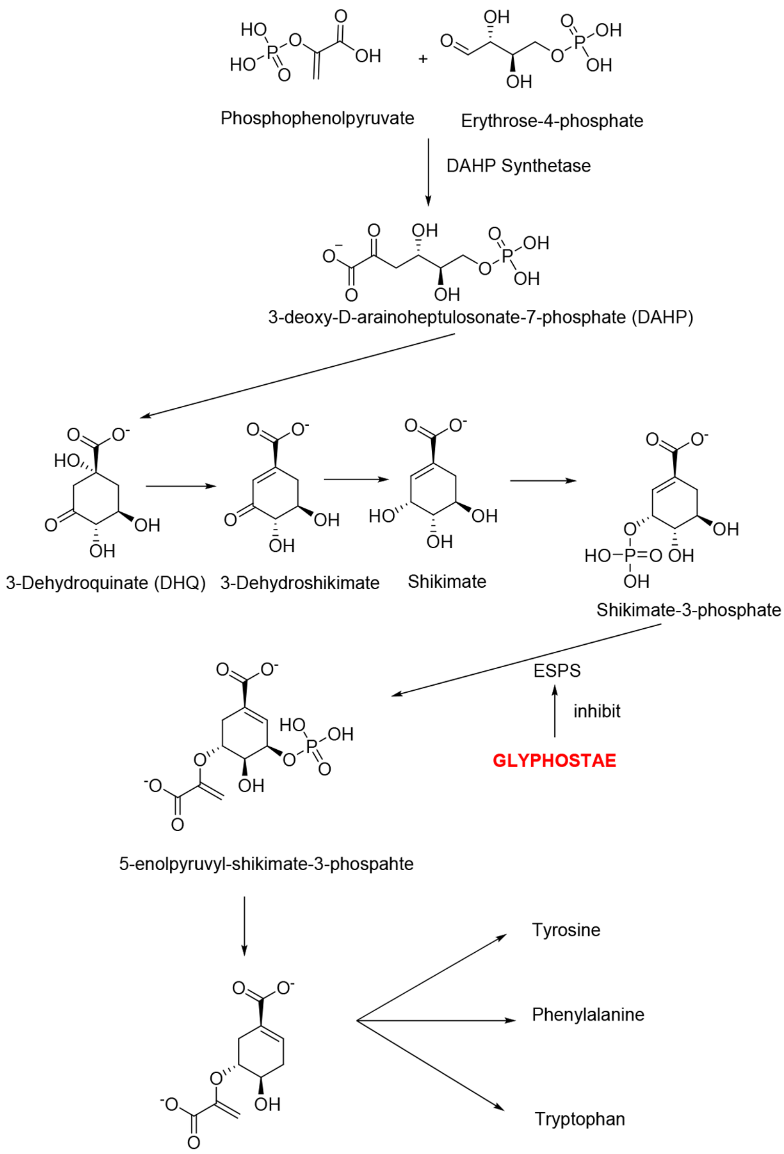

Figure 4. Schematic representation of the shikimate pathway: mode of action of glyphosate by inhibiting EPSPS (5-enolpyruvylshikimate-3-phosphate synthase).

2.4. Mode of Action. Glyphosate is a wide-spectrum herbicide considered as both toxicologically and ecologically safe that inhibits aromatic amino acid production by inhibiting the crucial enzyme in the shikimate pathway.[49] The plants, fungi, and bacteria possess this 5-enolpyruvylshikimate-3-phosphate synthase (EPSPS) enzyme, but it is lacking in animals.[50] With the introduction of glyphosate in the market, it acquired a dominant status in the pesticide market, which could be attributed to its mode of action and rapid translocation.[51] It is translocated well, and its action is gradual enough to take advantage of this rapid translocation. There are no competing herbicide analogues or classes, since glyphosate is the sole herbicide that targets the binding and inactivation of the crucial enzyme of the shikimate pathway, the process by which it destroys plants and microorganisms.[52] Further, EPSPS favors the formation of chorismate, a precursor for aromatic amino acids production in the form of tryptophan, tyrosine, and phenylalanine that participate in the shikimate pathway.[19] It hinders the enzyme 5-enolpyruvylshikimate-3-phosphate synthase, which initiates the reaction between shikimate-3-phosphate (S3P) and phosphoenolpyruvate for the formation of 5-enolpyruvylshikimate-3-phosphate (EPSP). It was determined that glyphosate is primarily absorbed by the leaves and only minimally through the roots, is efficient in rapid growing plants, and is unable to stop seeds from germinating.[53] The mode of action of glyphosate through the pathway is illustrated in Figure 4. After glyphosate application, it is readily taken up by the plants to growing roots and leaves, and this systemic action is crucial for its activity. As a result of enzyme inhibition, shikimate-3-phosphate is stored in plant tissues, diverting valuable resources and energy away from other functions and eventually killing the plant. It was found that plant growth ceases within hours of treatment, while the leaves take many days to become yellow. Glyphosate has the capability to chelate CO^{2+}, which contributes to its mechanism of action. As a result, the growth of several plants is hampered by glyphosate inhibitory activity,[54] and insufficiency of the enzyme activity results in a lack of aromatic amino acids, which in turn impacts

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Table 2. Bacteria Strains That Degrade Glyphosate

bacteria strain	bacteria source	degradation pathway	gram status	metabolites detected	comment	ref
Pseudomonas sp. PG2982	*Pseudomonas aeruginosa* ATCC 9027	sarcosine pathway	−	phosphate, glycine, sarcosine, formaldehyde	utilizing glyphosate as only phosphorus source	Jacob et al. (1985),[60] Kishore and Jacob (1987)[65]
Arthrobacter sp. GLP-1	*Klebsiella pneumonia* accidental contaminant	sarcosine pathway	+	phosphate, glycine	utilizing glyphosate as only phosphorus source	Pipke et al. (1987)[66]
Agrobacterium radiobacter	Wastewater from an American water treatment	sarcosine pathway	−	data not available	utilizing glyphosate as only phosphorus source	McAuliffe et al. (1990)[67]
Arthrobacter sp. GLP-1/Nit-1	*Arthrobacter* sp. GLP-1/Nit-1 is a mutant of *Arthrobacter* sp. GLP-1	sarcosine pathway	+	phosphate	utilizing glyphosate as both phosphorus and nitrogen source	Pipke and Amrhein (1998)[62]
Streptomyces sp.	Municipal sewage treatment plants untreated sludge	sarcosine pathway	+	glycine, sacrosine	utilizing glyphosate as phosphorus, nitrogen, and phosphorus source	Obojska et al. (1999)[68]

the different metabolic processes of plants. These amino acids are necessary for the formation of proteins that connect primary and secondary metabolites. Glyphosate can also function as an antagonistic inhibitor of phosphoenolpyruvate (PEP), which is a precursor to the production of aromatic amino acids. It also has an influential activity on other metabolic processes, which may be crucial in glyphosate's overall fatal activity.[55]

4. DIFFERENT METHODS OF GLYPHOSATE DEGRADATION

Glyphosate is sensitive to breakdown by both chemical reactions and microorganisms in the environment. In addition, glyphosate undergoes the process of photodegradation.

4.1. Chemical Decomposition. Glyphosate does not readily oxidize or get hydrolyzed after it has been applied to fields. Yet, the decomposition of glyphosate follows pseudo-first-order dynamics when it is carried out in water. It is because the degradation of glyphosate in water is accomplished through the utilization of oxidatively accelerated processes. At 20 °C, an aqueous suspension of birnessite catalyzes the abiotic degradation of glyphosate and its most widespread metabolite, AMPA (a mineral of Mn along with Ca, K, and Na present commonly in the soils).[56] The process of abiotic breakdown occurs on the outermost layer of manganese oxide and includes the breakage of the C−P bond. The removal of *N*-phosphonomethyl glycine by manganese oxide occurs more quickly and is accelerated by an increase in temperature. Since copper ions are present, the degradation pathway was able to function less efficiently. Copper ions form a strong coordination complex with the glyphosate molecule, which in turn limits the binding of glyphosate to the reactive oxidation sites on the surface of the manganese oxide.[57]

4.2. Microbial Degradation. Glyphosate is adsorbed due to complex formation between its phosphonate group and soil-exchanged polyvalent cations, and its adsorption is influenced by parameters such as organic carbon content, clay content, cation-exchange capacity, and soil pH. Since glyphosate is firmly bonded to soil and exhibits a minimal desorption rate (approximately 5−24% of the initially adsorbed glyphosate), a very small quantity of glyphosate remains accessible inside the soil for microbial breakdown and utilization by the plant. The glyphosate median half-life period has been described 2−91 days in aquatic media, whereas it is 2−215 days in soil.[58] As a result of the fact that only 5−24% of originally sorbed glyphosate is desorbed and that left out remains confined within the soil, only a trace quantity of glyphosate is left accessible in soil for plant absorption, breakdown, and interaction.[16] Glyphosate microbial degradation depends on type of microbe community of each soil type.[59] Because of the breakage of the C−P bond, glyphosate can be readily degraded by enzyme reactions of microorganisms. An example of this kind of metabolic pathway can be seen in the *Pseudomonas* PG2982 culture, which converts glyphosate into phosphorus.[60] Various different microorganisms such as *Rhizobium meliloti*, other *Rhizobium* strains, *Agrobacterium radiobacter*, and the *Arthrobacter* GLP-1 strain that also show analogous pathways that degrade glyphosate[61] are discussed below in Table 2. In the case of *Arthrobacter* strain GLP-1, the pathway was observed in a mutant known as *Arthrobacter* GLP-1/Nit-1, where glyphosate was employed as a source of nitrogen.[62] Glyphosate is utilized by *Streptomyces* species as a source for both including nitrogen and phosphorus.[63] The glyphosate metabolite aminomethylphosphonic acid (AMPA), which is formed during microbial degradation, is non-toxic and degrades at a slower rate than glyphosate itself. As phosphorus is an essential component for bacteria, the quantity of phosphorus in glyphosate renders it significantly susceptible to the breakdown process facilitated by microorganisms.[64]

4.3. Photodegradation. Glyphosate is also receptive to photodegradation, and it has a four day half-life in deionized water under exposure to ultraviolet light. Nevertheless, glyphosate does not undergo any photodegradation when exposed to long-wavelength light.[69] Glyphosate has the potential to be broken down by ultraviolet light into aminomethylphosphonic acid, which is resistant to the deteriorating effects of light.[70]

5. GLYPHOSATE IMPACT ON LIVING ORGANISMS

5.1. Impact on Human Health. The widespread application of glyphosate in the past years has raised issues regarding the herbicide's potential toxicity and its repercussions for human health. As a result, research into how glyphosate affects human health has grown. Due to the existence of the shikimate pathway, glyphosate had fatal effects on plants but was less harmful to humans and other animals at even high concentrations. For terrestrial and aquatic animals, the concentration of glyphosate dosage ranges from 175 to 540 mg and from 1 to 53 mg, respectively.[71,72] Although glyphosate-based herbicides are not easily absorbed via the skin in humans, they are consumed indirectly through the consumption of crops that have been treated with them. The accumulation of glyphosate-based formulations found in urine and faeces that do not degrade into other substances is a significant health hazard caused by glyphosate absorption or ingestion.[73] Further, it can lead to toxicity in humans, which is normally categorized into two forms. Acute toxicity is described as the risk associated with a chemical's vulnerability

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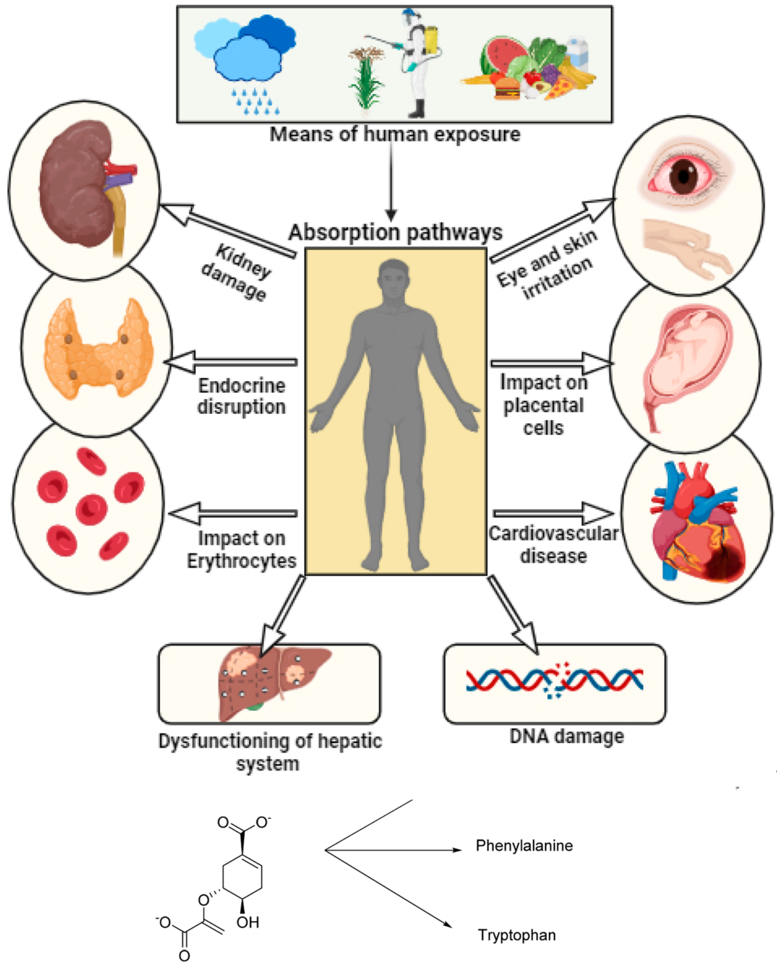

Figure 5. Means of glyphosate exposure and various impacts of glyphosate on human health.

as a result of inhalation, skin contact, and oral contact at the time of spraying the chemicals. Chronic toxicity referred to the risk brought by consuming small amounts of substances continuously through diet and hazards related with it.[74] With glyphosate, the acute LD50 (lethal dosage, 50%) in terms of mammalian toxicity is estimated to be around 5037 mg kg^{-1}. According to the EPA registration, any herbicide with an acute LD50 greater than 5000 mg kg^{-1} will come under category IV, which has the lowest acute toxicity. Glyphosate's LD50 levels are almost on the threshold of being classified as a category IV chemical.[75]

With the increase in glyphosate use, its excessive concentrations result in health problems like cancer, kidney damage, attention deficit hyperactivity disorder (ADHD), autism, Parkinson's, and Alzheimer's diseases, as well as an increase in eye and skin irritation.[76−78] Glyphosate ingestion and remains on vegetables, fruits, and animals, as well as a surge in the use of genetically modified (GM) crops such as grains, soybeans, and maize, are two major ways through which diet exposes the general population to glyphosate. It also has an impact on pregnant and lactating mothers who are exposed to glyphosate. These factors can result in preterm or premature births (births that take place prior to 37 weeks of gestation). Furthermore, the traces of glyphosate in preterm birth raise the threat of infectious diseases, respiratory failure, feeding problems, intraventricular hemorrhage, sepsis, and long-term disorders such as cardiovascular disease, nephropathy, neural development complications, and respiratory dysfunction, as illustrated in Figure 5.[79] Over the years, various studies have been conducted in the past explaining serious implications on human health, and one of them revealed the risk of celiac disease and gastrointestinal problems on exposure to glyphosate.[80] Continuous exposure to glyphosate and glyphosate-based herbicides at high concentrations produced arrhythmias and cardiotoxic effects through anomalies in the electrocardiogram (ECG).[81,82] Additionally, glyphosate and GBH increase the amount of oxygen free radicals, causing oxidative stress and thereby damaging the organs,[83] whereas low concentrations were found to cause hepatotoxicity in rats upon chronic exposure as well as dysfunctioning of the hepatic system.[84] In 2019, an investigation revealed that chronic exposure could lead to nephrotoxicity,[85] renal diseases, and chronic kidney disease.[86]

Human cells were used to assess the cytotoxicity of glyphosate and glyphosate-based herbicides. GBHs were found to cause morphological alterations in human erythrocytes in one research.[87] Another investigation using nerve cells, liver, and lungs observed that there is potential of cytotoxicity linked with GBHs; however, this might be due to glyphosate-based herbicide elements other than glyphosate.[88] Various government and international institutes have conducted carcinogenic studies on glyphosate in recent years.

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Table 3. Various Side Effects Associated with Glyphosate Application

side effect	description	refs
skin irritation	direct contact with glyphosate formulations can cause skin irritation or dermatitis in some individuals	Acquavella et al. (2016)[98]
eye irritation	contact with the eyes can lead to irritation, redness, and discomfort	Williams et al. (2000)[54]
respiratory issues	inhalation of glyphosate spray mists can cause respiratory irritation, coughing, or difficulty breathing	Northall et al. (1999)[99]
gastrointestinal discomfort	ingestion of significant amounts can lead to nausea, vomiting, or diarrhea	Priyanto et al. (2020)[100]
allergic reactions	some people may experience allergic reactions, though they are relatively rare	Ndlovu et al. (2011)[101]
developmental effects	concerns have been raised about potential developmental effects, especially in fetuses and young children	de Araujo et al. (2016)[102]
environmental impact	glyphosate's use has been associated with ecological impacts, including harm to nontarget plants and animals	Cerdeira et al. (2006)[103]
resistance in weeds	prolonged use of glyphosate has led to the development of herbicide-resistant weeds, impacting agriculture	Gage et al. (2019)[104]

Nevertheless, this assessment does not reflect scientific consensus, and there is currently a heated debate about the status of glyphosate as a carcinogen in nature.[89] Glyphosate was recognized as a possible human carcinogen and placed in group 2A by the International Agency for Research on Cancer (IARC) in 2015. This categorization is determined based on limited proof of human carcinogenicity (studies depicted positive correlation among glyphosate and non-Hodgkin lymphoma (NHL)) and adequate information in animal medical research.[90,91] The Food and Agriculture Organization (FAO) and the World Health Organization (WHO) produced a joint assessment on pesticide residues in food in 2016, concluding that people exposed to glyphosate through diet are unlikely to exhibit carcinogenesis and hence glyphosate is not a carcinogenic in nature.[92] In 2017, both the European Chemical Agency (ECHA) and European Food Safety Authority (EFSA) reached the decision that glyphosate was not carcinogen in nature.[93] However, in a study on the toxicity of glyphosate in 2019, the U.S. Agency for Toxic Substances and Disease Registry (ATSDR) established that there may be a carcinogenicity linked with usage of glyphosate and GBHs.[94,95] The latest research reported in 2019 found a higher risk of NHL in people who were subjected to GBHs heavily;[96] however, according to a review on epidemiological data published in 2020, there is no correlation between glyphosate exposure and the development of NHL.[97] Table 3 depicts the various side effects associated with glyphosate application.

In vitro investigations in human cells have demonstrated that a low daily dose of glyphosate can impair the activity of acetylcholinesterase, resulting in dysregulation of nerve impulse transmission and the emergence of neurological diseases.[105] However, animal studies indicate that glyphosate is not neurotoxic even at high doses, thus various organization such as EFSA and FAO do not classify it as neurotoxic in nature.[106,92,107] In a series of in vitro studies conducted, glyphosate and GBHs damaged human cells, mammalian chromosomes, and DNA; as a resul,t IARC designated glyphosate as a genotoxic substance in 2015.[90,91] In later years, it was observed that glyphosate has no genotoxicity in humans according to the reports published by a number of international organizations, including EFSA and FAO.[106,92] An in vitro study published in 2019 revealed that glyphosate could cause epigenetic alterations in animal cells and DNA damage in human leukocytes.[108] According to a systematic review that was released in 2019, exposure to GBHs has a genotoxic effect. Nevertheless, polyethoxylated tallow amine (POEA) surfactant found in GBHs may be the possible cause of genotoxicity rather than glyphosate.[109]

5.2. Impact on Animals. Glyphosate exhibits adverse effects on both unicellular and multicellular organisms established in water and soil. In unicellular organisms, glyphosate impacts single-celled simple organisms such as *Euglena gracilis* by reducing their photosynthesis, chlorophyll concentration, and respiration.[110] It also affects mycorrhizal fungal species such as *Laccarialaccata*, *Hebelomacrustuliniforme, T. terrestris*, *Suillustomentosus*, and *Thelephora Americana* by reducing their growth.[111] Glyphosate affects soil rhizosphere-related communities by increasing the relative abundance of γ-proteobacteria.[112] In vitro studies were carried out to analyze glyphosate effects on harmful bacteria such as *Salmonella typhimurium*, *Salmonella enteritidis*, *Salmonella gallinarum*, *Clostridium botulin*, and *Clostridium perfringens*. It was observed that these pathogenic bacteria are glyphosate-resistant.[113] Further, glyphosate also affects the periphyton communities including lentic as well as lotic systems by reducing rates of photosynthesis and changes in the structure of the diatom community, respectively.[114,115] In multicellular organisms, the toxicological effects of glyphosate and its commercial formulation Rondo were studied using two freshwater algal species, namely, *Scenedesmus quadricaud* and *Scenedesmus acutus*, and it was seen that no acute toxicity was caused to these algal species.[116] In invertebrates, Nemathelminthes are parasitic lower invertebrates that impact ecosystems and contribute to the nutrient cycle. Glyphosate use in soil systems has infected nematodes and caused harmful effects. A study by researchers found that glyphosate concentrations ranging from 0.1 to 8 mg/L significantly reduced nematode *Chordodesnobili* and the infective ability in larvae. Glyphosate of both technical grade and formulation provided identical outcomes.[117] In the phylum Annelid, as we know earthworms are a vital element of soil vegetation as they help preserve the quality and fertility of the soil ecosystem.[118] In vitro studies were carried to investigate the toxicological effects at various concentrations of glyphosate on *Eisenia fetida*, and no death rate was observed; however, a consistent and significant reduction in the average body mass was recorded at every test doses. Adverse toxic effects on breeding and earthworm's growth were also observed. There were no cocoons or young discovered in the herbicide-treated soil. Important anatomical alterations were also noticed after running the trial for 30 days. When subjected to the highest test dose of glyphosate-treated soil, all samples showed morphological anomalies such as elevated bodies, coiling, and curling.[119] Another research team examined the acute and long-term toxicological effects of glyphosate's primary metabolite, aminomethylphosphonic acid, on *Eisenia andrei* at field-relevant doses. Both acute and chronic tests showed no significant mortality. When earthworms were subjected to an acute toxicity test, the amino-

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Table 4. Glyphosate effect on both Invertebrates and Vertebrates

phylum	species	treatment duration	outcome	ref
Nemathelminthes	*Chordodes nobili*	96 h	reduced infective ability in larvae	Achiorno et al. (2008)[117]
Annelida	*Eisenia fetida*	30 days	average body mass reduction, no cocoons or young observed	Correia and Moreira (2010)[119]
Arthropoda (Crustacea)	*Daphnia magna*	55 days	reduced juvenile size, growth, fertility, and abortion rates	Cuhra et al. (2013)[121]
Arthopoda (Insecta)	*Apis mellifera*	14 days	acetylcholinesterase activity decreased	Boily et al. (2013)[123]
Mollusca	*Helix aspersa*	168 days	snail tissues detected with glyphosate residues	Druart et al. (2011)[124]
				Annett et al. (2014)[125]
echinoderms	*Sphaerechinus granularis*		cell cycle hampered, embryos developing into abnormal adults	Marc et al. (2004)[126]
fish	*Pimephalespromelas, Salmo gairdneri, Ictalurus punctatus, Lepomis macrochirus*	96 hours	fishes' life cycle impacts the herbicides toxicity effects.	Folmar et al. (1979)[127]
				Stanley et al. (2016)[128]
amphibians	*Lithobates catesbeianus*	14 days	decreased glycogen and lipid levels, increased lipid peroxidation, cholesterol in gills, while muscle and protein content decreased	Dornelles and Oliveira (2016)[129]
				Dornelles and Oliveira (2014)[130]
reptiles	*Salvator meriana*		dna damage on erythrocytes, affects neonate's development	Schaumburg et al. (2016)[131]
				Freitas et al. (2020)[132]
birds	*Anas platyrhynchos*	15 days	reproductive system damaged, affecting androgen and estrogen synthesis, testis and epididymal morphology, male genital organs	Oliveira et al. (2007)[133]
				Cassault-Meyer et al. (2014)[134]
mammals	Wistar Rats	21 days	male rats experienced reduced sperm content, defective sperms, and decreased testosterone levels	Dallegrave et al. (2007)[135]
				Paiva et al. (2021)[136]

methylphosphonic acid-treated earthworms had a dramatic reduction in biomass. However, at the highest dose of aminomethylphosphonic acid used in the chronic test, a greater reduction in the biomass of earthworms was observed. The quantity of young animals and cocoons also increased where the herbicide concentration was the highest. Yet, it was observed that these juveniles' mean body weight had reduced. It was confirmed with these results that adults are less susceptible to aminomethylphosphonic acid than juveniles.[120]

Under the phylum Arthropoda and subphylum Crustacea, research carried out on the effects of glyphosate and Roundup on *Daphnia magna* found that both herbicides are hazardous. Roundup had slightly lower acute toxicity than pure glyphosate but had higher chronic toxicity. It reduced juvenile size, growth, fertility, and abortion rates at concentrations of 0.05 mg/L. Both herbicides had significant unfavorable effects at doses of 1.35 and 4.05 mg/L.[121,122] In insects, researchers studied acetylcholinesterase activity in honeybee species *Apis mellifera* exposed to maize and subjected to treatment with neonicotinoids, glyphosate, and acephate. They found abnormally high levels in neonicotinoid-treated bees and slightly lower levels in glyphosate-treated bees.[123] In molluscs, researchers assessed the effects of glyphosate over a prolonged period of time on a type of terrestrial snail called *Helix aspersa*. They exposed young, emerging snails to soil and food that had been polluted with the pesticide for 168 days. No effects on survival or growth were observed. However, glyphosate was detected in snail tissues fed contaminated food, suggesting a potential food chain risk.[124,125] In echinoderms, researchers investigated the impact of glyphosate-based herbicides on sea urchin cell division. Various glyphosate formulations with different doses were employed and it was determined that glyphosate at a maximum dose of 2 mM stopped the cell cycle, resulting in the embryos developing into abnormal adults.[126] Under vertebrates, the researcher tested the toxicity on four different freshwater fish, including channel catfish (*Ictalurus punctatus*), rainbow trout (*Salmo gairdneri*), bluegills (*Lepomis macrochirus*), and fathead minnows (*Pimephalespromelas*), using the Roundup surfactant, formulated herbicide Roundup, technical grade glyphosate, and isopropyl amine salt of glyphosate. According to an acute toxicity test, Roundup's LC50 value at 96 h differed among different fish species; for example, the LC50 at 96 h for fathead minnows was 2.3 mg/L, but that for rainbow trout was 140 mg/L. Surfactant toxicity was shown to be comparable to Roundup formulation toxicity. However, it was discovered that Roundup was considerably more harmful to rainbow trout and bluegills at a pH of 7.5 and higher temperatures. The fish's life cycle impacts the herbicide's toxicity effects. Sac fry and early swim-up stages are more susceptible to glyphosate, whereas eyed eggs were a less delicate stage. This herbicide had no effect on adult rainbow trout's gonadosomatic index or reproduction.[127,128] In amphibians, researchers found that herbicides such as atrazine, glyphosate, and quinclorac significantly affected bull frog

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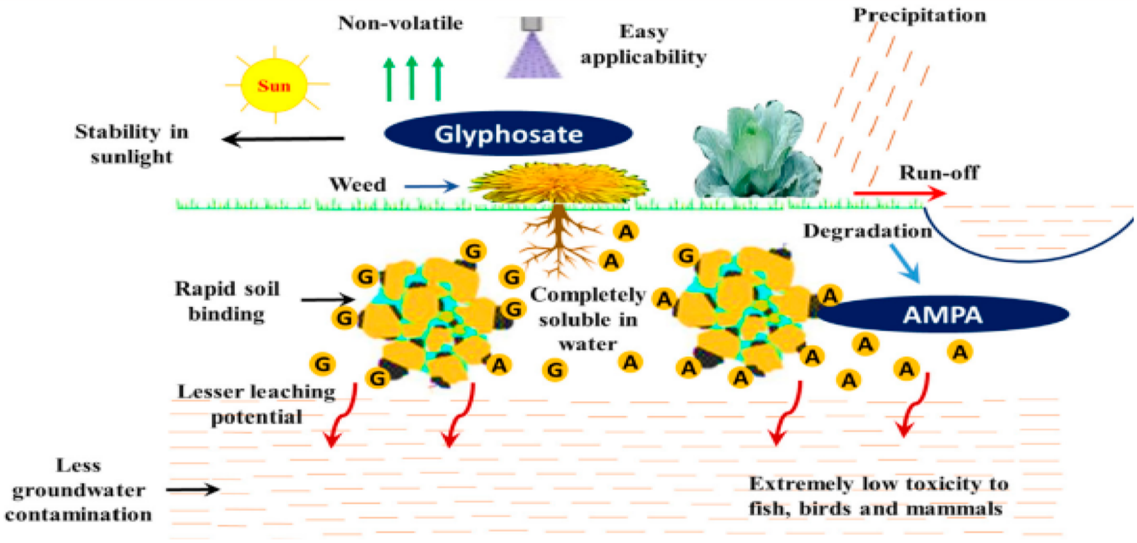

Figure 6. Environmental variation in bioavailability and toxicity of glyphosate in different abiotic spheres. Reproduced with permission from ref 87. Copyright 2020 Elsevier.

tadpole endurance and biochemical changes. Glyphosate exposure led to decreased glycogen and lipid levels and increased lipid peroxidation and cholesterol in gills, while muscle and protein content decreased.[129,130] In reptiles, researchers found that the most frequently employed herbicide, Roundup, has genotoxic adverse effects on tegu lizards, producing DNA damage on the red blood cells and potentially affecting neonate development. The study revealed potential risks to the reptile's health.[131,132] In birds, researchers analyzed in vivo studies that indicate that glyphosate-based Roundup has severe effects on the male drake *Anas platyrhynchos* reproductive system, affecting androgen and estrogen synthesis, testis and epididymal morphology, and male genital organs. The reproductive system of the species is negatively impacted by these effects, which are dose-dependent.[133,134] In mammals, a study examined the reproductive systems of Wistar rats exposed to glyphosate and polyoxyethyleneamine, the active ingredients in the herbicide Roundup. Female rats showed no maternal harm, while male rats experienced reduced sperm content, defective sperm, and decreased testosterone levels.[135,136] Various numerous effects of glyphosate are depicted in Table 4 below for both invertebrates as well as vertebrates.

6. GLYPHOSATE'S IMPACT ON THE ENVIRONMENT

6.1. Occurrence. Humans are constantly exposed to the glyphosate residues through various routes such as soil, water and air.[137] The persistence of glyphosate in the environment is due to its strong adsorption on soil particles causing its transmission into humans ultimately by food through wind and water erosion.[138] The anthropogenic degradation of aquatic habitats by the excessive use of pesticides has come up as a condemning environmental issue. These harmful synthetic chemicals find their path into water bodies through air/water interaction, deposition in atmosphere, agricultural runoff, and effluent discharge.[139] Owing to the short lifetime of glyphosate, the molecule was rapidly degraded by aquatic organisms and soil, reducing environmental pollution.[140] However, its residues and AMPA could enter the dissolved phase in groundwater, leading to contamination via vertical transportation through the soil profile.[141] Seasonal fluctuations in the glyphosate concentrations in water were noticed, which could be attributed to combined factors such as climatic cycles, hydrology, application of pesticide, land management, and biodegradation.[142,143] Moreover, the airborne impact of glyphosate remains a promising concern. The remains of pesticide droplets that evaporate before hitting the target and wind-blown soil particles can cause the pesticides to drift to nontarget organisms in the vicinity.[144] Due to the low vapor pressure and its facile solubility in the water, it easily dissolves in the dew drops in high humid environment.[145] The concentration of pesticide in the environment is associated with various factors like temperature, land use, and radiation, which also affect its presence.[146] Further, the presence of glyphosate in the lithosphere possesses serious implications for environment. The high binding affinity of the herbicide mostly founds its accumulation on the top layer of the soil, thereby restricting its mobility to a considerable extent.[147] The pH plays a significant role in the adsorption of glyphosate to soil particles. At pH 4−8, glyphosate adsorption increases, while with decrease in pH the ligand transformation into amorphous iron and aluminum oxide is facilitated.[148] Glyphosate is also found in the lower layers of the soil profile, which implies that its adsorption is not perpetual and presence can be detected post four months of applications.[149] Its persistence depends on a biotic component, climatic conditions, physiochemical properties, and soil composition.[150] The high water solubility of glyphosate and its metabolites is indeed a crucial factor contributing to the lack of bioaccumulation or biomagnification in organisms within natural ecosystems.[151] Overall, the widespread and prolonged use of glyphosate and other pesticides has emerged as a serious environmental nuisance, highlighting the need for better monitoring and management practices in agricultural grounds to minimize the catastrophic impact on environment.

6.2. Resistance in Weeds. Plants of higher taxa exhibit vulnerability toward glyphosate and are not naturally resistant to it. Certain plants are more vulnerable than others due to their physiological and biochemical characters. Glyphosate has proven to be more resistant to weeds such as Bermuda grass

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(*Cynodon dactylon*) and field bindweed (*Convolvulus arvensis*).[152−154] Various weed species and plants are showing resistance due to widespread usage of glyphosate globally, and first resistance was noted in Australia crops of annual ryegrass (*Lolium rigidum*).[155] Glyphosate resistance crops are widely cultivated in significant regions of the United States, Argentina, and Brazil, which has also led to the discovery of a small number of weeds that are glyphosate-resistant. The previously identified ragweed (*Ambrosia artemisiifolia*), glyphosate-resistant horseweed (*Conyza canadensis*), and lambsquarters (*Chenopodium album*) are often widespread in the United States.[156]

Crops such as corn, cotton, and soybeans that exhibited resistance to glyphosate were first introduced to the market in 1996 as a direct result of the widespread application of glyphosate in the agricultural sector.[157] Crops grown across the world[158] and CP4 genes of an *Agrobacterium tumefaciens* species have been utilized to transcribe glyphosate-resistant EPSPS. The CP4 genes and a promoter were inserted into the genomes of crops that exhibit elevated amounts of glyphosate resistance. In the case of maize, mutagenesis of maize genes was done to produce glyphosate-resistant 5-enolpyruvylshikimate-3-phosphate synthase.[159,160] In farming, the utilization of modified transgenic crops has encouraged unrestricted glyphosate use magnified by numerous times, causing cropping systems to deteriorate. It has become difficult to suppress the weeds as now they have become more resistant to this herbicide.[161−164] Glyphosate has become a significant constituent of most water bodies and soil systems as it has been used in larger amounts over the years.[55] The resultant metabolite, AMPA, is now observed in numerous food crops during harvest and even in processed food. Overall, glyphosate alone and its different breakdown constituents are present in ecological systems, as depicted in Figure 6, at virtually every food chain level and are exerting harmful impact over large range of species.[165]

Homes and co-worker compiled a list of "The World's Worst Weed" and also showed that four families are at significantly higher risk of resistance, i.e., Asteraceae, Poaceae, Amaranthaceae, and Chenopodiaceae, in comparison to other prominent weeds.[168] When compared to other significant weed families, Poaceae exhibited a higher activity for glyphosate. The fact that more grasses than any other family of weeds have developed resistance to glyphosate is interesting. While 25% of the major weeds in the globe are grasses,[106,166] it is estimated that glyphosate-resistant grasses account for 47% of all glyphosate-resistant weeds. As a result, the dose−response curve for glyphosate dosages on grass species is steeper than that on nongrass species. As compared to other weed families, Poaceae, which are mostly grasses, get more exposure. Poaceae family members are present in major continents and almost all regions of the world, which naturally makes them susceptible from glyphosate, leading to higher resistant individual weed varieties. Compared to the commonly held belief that low doses are more effective at selection for resistance, high dose rates more successfully yield glyphosate-resistant grasses. Eight of the 17 incidences of glyphosate resistance among Poaceae were found in the genera *Lolium*, *Chloris*, and *Bromus*. *Lolium* sp., *Digitaria insularis*, *Eleusine indica*, *Sorghum halepense*, and *Echinochloa colona* species with glyphosate resistance have the widest spread and enhanced economic impact. Weeds in the family Asteraceae make up to 30% of the glyphosate-resistant weeds and also constitute 16% of the world's worst weeds. Major genera from the family, including Conyza and Ambrosia express the resistant characteristics and are almost half of all resistant weeds in the family.

Amaranthus is the singular genus of 163 genera of the Amaranthaceae family that have developed glyphosate tolerance capability. The *Amaranthus* species *A. palmeri*, *A. tuberculatus*, *A. hybridus*, and *A. spinosus* have all developed glyphosate resistance. *A. tuberculatus* and *A. palmeri* are two species of significant economic importance, but both have developed resistance in the main herbicide sites of action that are employed to manage them. *Kochia scoparia* and *Salsola tragus* are two weed species that belong to the Chenopodiaceae family, and both have developed resistance to glyphosate. Due to its effective tumbleweed seed distribution strategy, glyphosate-resistant *K. scoparia* is currently a significant economic concern in the Great Plains of the United States and also in the Prairie Provinces of Canada. *S. tragus* is a tumbleweed and has recently been found to be glyphosate-resistant in small populations in Montana and Oregon.[167,32] It is interesting to observe if *Kochia*-like rapid expansion occurs by its unique dispersal mechanism.[169]

6.3. Next Generation and Future of Glyphosate Crops. The time when crops could only withstand glyphosate is coming to an end.[170,171] The development of "third generation" GR crops combines glyphosate features with those of other herbicides. The most popular strategy uses triple herbicide-resistant crops (HR crops) that are resistant to glufosinate, glyphosate, and other types of herbicide. The GR crop systems will be a combination of features that will allow new herbicide compositions to tackle weed resistance mechanisms.[172] The present developmental tactics of adding further resistance mechanism to GR crops does not deliver high recurrence for a dicey investment, despite years of protective use, and the channel of novel genetically modified HR crops may come to an end, which could be attributed to a slow and expensive regulatory process. The following generation of HR agricultural technology will be synthetic auxin 2,4-dichlorophenoxyacetic acid- (2,4-D) and dicamba-resistant.[173,174] New formulations with a lower amount of volatile salts and improvised application procedures are being touted as remedies to prevent off-target drift. Auxin-resistant soybeans and cotton will permit new uses of these old herbicides.[175] The problems of off-target shifting and impacts on nontarget organisms are so prevalent that Dow's assertion of synergism in a patent application gravely endangered the registration of the novel 2,4-D and glyphosate mixture.[176,177] The cost of the 2,4-D product could be significantly increased by the addition of the new inert chemicals; therefore, they must be effective. Crops that are resistant to herbicides that inhibit 4-hydroxyphenyl pyruvate dioxygenase (HPPD) will come after the auxin crops. Key weeds can be controlled by HPPD-inhibiting herbicides with some soil residual activity, but the spread of *Palmer amaranth* and water hemp before their commercialization will reduce the effectiveness of this technology.[178] Two features that will allow some HPPD herbicides to be used in new ways are currently being developed.[179,180] Corn typically has some built-in resistance to HPPD herbicides, similar to auxin herbicides; thus, soybeans and cotton will be the crops where the technique will be most useful. Additionally, there will be commercially available HR crops modified to be resistant to acetolactate synthase (ALA) and acetyl coenzyme A carboxylase (ACCase) inhibiting herbicides. Despite considerable weed resistance, these control

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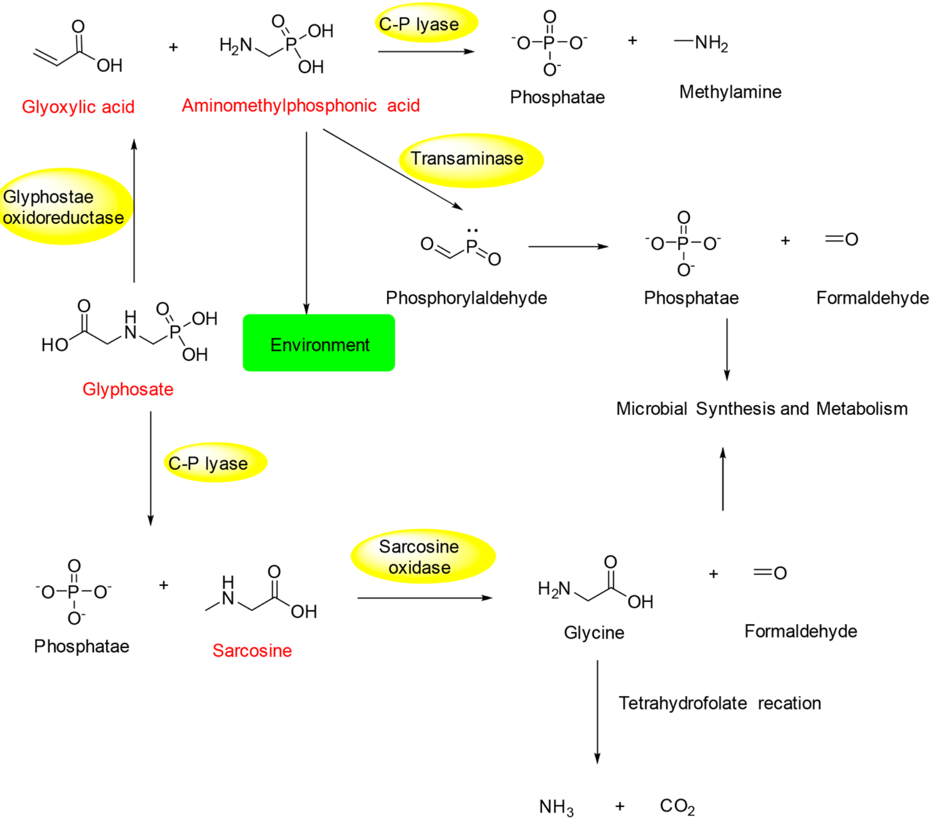

Figure 7. Scheme illustrating the degradation mechanism of glyphosate via different pathways.

techniques are still moderately effective.[181,182] Various defense strategies against herbicides might be dependent on metabolic degradation by transgenic cytochrome P450 and glutathione-*S*-transferase (GST) enzymes. Alternatively, weeds have developed a comparable nontarget site resistance phenomenon, which would probably reduce the usefulness of these strategies.[183,184] Despite having received widespread approval from numerous national regulatory agencies, glyphosate and GR crops continue to be the focus of daily news reports regarding possible health hazards. 2015 saw a resurgence of health concern, as the International Agency for Research on Cancer (IARC) released a monograph stating that the herbicide is "probably carcinogenic to humans".[185] Despite the fact that the U.S. Environmental Protection Agency and others have recently come to conclusion that the herbicide is probably not carcinogenic,[186] the debate about its protection persists in both the press and academic journals.[187−189]

Furthermore, questions are frequently raised concerning the safety of the herbicide formulations using the original tallow amine ethoxylate "Roundup surfactant". This surfactant type has emerged with marine and eye toxicity issues,[6,190] which are generally handled with obligatory application limitations and operator personal protective equipment (PPE) requirements. The significant level of civic concern about the herbicide and GM crop safety guidelines are projected to persist, leading to glyphosate bans in some areas and the continued prohibition of GM crop production in much of Europe.[191] These implications are unlikely to diminish the gross amount of herbicides used in GR crops around the world. The herbicide will remain the backbone of weed management, even once the glyphosate trait is integrated into numerous HR crops. Despite its shortcomings, glyphosate remains effective against most unwanted plants, and sales are expected to grow tremendously. The emergence of such weeds provides another motivation for industries to restart efforts against herbicide discovery. The discovery of a new herbicide class and its related trait could be very expensive, but it would also be perilous because of its resistance against nontarget organisms and the cross-resistance phenomenon, which limits its utility as a new herbicide, making it ineffective for such extravagant and decade-long development process. Furthermore, no pesticide could provide a permanent solution, since they could produce resistance. The failure to discover a mode of action over the last three decades is a major source of concern. Despite its shortcomings, glyphosate remains effective against most weeds, and sales are expected to reach $10 billion by 2021.[192] The anticipated mergers of some enormous pesticide businesses should ensure for the continuation of several herbicide discovery programs, and the lingering megacompanies should fund a viable endeavor with research support founded on sales proportion. The dearth of discovery success has not yet resulted in a decrease in the chemical herbicide business. Ironically, the absenteeism of new herbicides has often resulted in growers paying more and employing older, virtually elapsed herbicides to combat HR weeds, resulting in what has been labeled as crop protection revival.[193] Co-development of a broad-spectrum herbicide and related HR crops could be extremely valuable and aids in revitalizing the herbicide trait business model. However, that technique is fraught with danger, and the expenditure required to concurrently commercialize a synthetic chemical and a biotech characteristic is likely to exceed 12 years and $500 million.[194,195] With the current pace of resistance development in some weeds, a sole new herbicide would be insufficient and would shortly lose its efficacy. If this

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is correct, the economics of these projects will fail, which may explain why firms have diverted money away from transgenic HR crops and toward less controlled technologies, including gene editing and ribonucleic acid interference (RNAi). Weed control strategies based on new chemicals and genetically engineered crops may already be too sluggish and expensive. For most growers, the current status quo of utilizing all existing weed managing strategies, chemical and nonchemical, by "throwing the kitchen sink" at HR weeds will work most of the time for a while, hopefully long enough to survive until researchers come up with new ideas. In the meantime, the lapse of over three decades without the unearthing of a novel chemical herbicide mode of action and the disastrous epidemic of resistant weeds are compelling growers to upsurge spending on weed management, creating a worst-of-both-worlds situation for the crop protection and seed industries.

6.4. Fate of Glyphosate in the Environment. The extensive application of glyphosate in agricultural farmlands has aroused concerns claiming its degrading impact on environment.[196] Degradation of glyphosate generally takes place via two enzymatic routes, as illustrated in Figure 7. One is through glyphosate oxidoreductase (GOX), while the other route is through carbon−phosphorus (C−P) lyase. The former path generally produces glyoxylate, a common natural metabolic compound and AMPA (which enters into environment through glyphosate degradation and detergents containing phosphorus), while the latter pathway produces sarcosine and inorganic phosphate.[197,198] The degradation rate is much higher in aerobic soil in comparison to that in anaerobic soil. Exchangeable soil acidity, ammonium-lactate-extractable potassium, and exchangeable calcium(III) ions are all strongly linked with the degradation of glyphosate.[199] Further, the presence of AMPA in soil shows more persistence than glyphosate, requiring the C−P lyase enzyme for degradation. The greater persistence of AMPA than glyphosate in soil may suggest that microorganisms that employ this breakdown route are less prevalent than those with GOX if this is the case for other microbes with C−P lyase. Microbes with such a C−P lyase can only obtain phosphorus from glyphosate or AMPA.[200] Plants can also metabolize glyphosate.[201] Plants can engage in both GOX and C−P lyase-mediated biological activities, but GOX metabolic glyphosate degradation is more common. Between species, glyphosate-induced AMPA synthesis varies greatly. No definitive generalization can be made, however;[202] legumes seem to be more capable in metabolizing glyphosate than grasses. At typical application rates, a significant amount of glyphosate absorbed by GR soybeans and canola is converted to AMPA.[203] Due to the total resistance of these plants to glyphosate at these application rates, the mechanism for degradation is unaffected by glyphosate phytotoxicity's secondary effects.[204] Most glyphosate-sensitive plants will only digest a modest quantity of glyphosate to AMPA because a substantial dose of glyphosate will likely impair metabolic function or cause the plant to die before much of glyphosate can be destroyed. Further, it is anticipated that in the future weeds with higher capacity for glyphosate metabolism could be found, as the majority of species have gene-encoded enzymes for glyphosate metabolism. This could gradually lessen the glyphosate's environmental half-life.

Since glyphosate breakdown occurs in plants, glyphosate and AMPA residues could be identified in plant products.[105] According to certain findings, glyphosate and its metabolites can also migrate through soil erosion caused by water and wind.[205] Both AMPA and glyphosate residues might last inside the soil for as long as six months, depending on the environment and soil where GBHs are sprayed.[206] Ecosystems may be impacted by glyphosate's ability to remain in the environment for several months.[19] According to a study, glyphosate alters the structure of soil microbes, which leads to the growth of phytopathogenic fungi.[23] The ability of glyphosate, on the other hand, to pollute aquatic ecosystems has been identified,[207] leading to the ECHA's classification of glyphosate as harmful to marine life with long-lasting consequences in 2017.[93]

7. CONCLUSION AND FUTURE PERSPECTIVE

Frequently considered an ecologically safe pesticide, glyphosate's indiscriminate application has put adverse implications on the environment as well as to humans. Exposure to glyphosate, the active ingredient in many herbicides, can indeed lead to numerous health effects depending on the route and amount of exposure. Some of the key health risks are skin irritation or dermatitis, eye irritation, respiratory issues upon inhalation, and gastrointestinal problems following ingestion. The possible developmental effects, particularly in fetuses and young children, have gathered attention and concern. Studies and discussions have focused on the possibility of adverse effects on development due to exposure, although the scientific consensus on this matter remains debated and evolving. Increased glyphosate usage causes serious concerns with regard to the herbicide's toxicity and the potential harmful consequences to nontargeted organisms. Presently, in the scientific community, no consensus about the glyphosate toxicity exists, specifically in relation to the herbicide's possible carcinogenic potential. Hence, extensive research is required to assess the toxicity of glyphosate as an active ingredient, particularly work on glyphosate-based herbicides, because the formulation may contain toxicants that can cause harm to humans. The development of resistance in weeds toward glyphosate, the active ingredient in herbicides like Roundup, has been a major focus of research in agriculture. Understanding the physiological mechanisms underlying this resistance is essential for properly controlling and effectively addressing weed resistance challenges effectively. One potential mechanism involves an increased level of 3-deoxy-D-arabino-heptulosonate 7-phosphate synthase (DAHPS), which is the primary enzyme in the shikimate pathway. The shikimate pathway is crucial for the synthesis of aromatic amino acids in plants. Glyphosate disrupts this pathway by inhibiting the enzyme 5-enolpyruvylshikimate-3-phosphate synthase (EPSPS), which leads to the impairment of amino acid production and ultimately causes plant death.

In glyphosate-resistant weeds, it is hypothesized that the enhanced activity or levels of DAHPS might contribute to the plants' ability to overcome the inhibitory effects of glyphosate. This increase in DAHPS activity could potentially facilitate enhanced carbon flow through the shikimate pathway, allowing the plants to maintain adequate levels of aromatic amino acids despite the presence of glyphosate. By having higher levels of DAHPS, resistant weeds might compensate for the inhibition of EPSPS induced by glyphosate, thereby supporting the production of essential amino acids and maintaining crucial metabolic processes. This elevated carbon flow through the shikimate pathway could be a part of the resistance mechanism, enabling the weed to survive glyphosate

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application. Research in this particular area assists scientists and agricultural experts in comprehending how weeds adapt and develop resistance to herbicides such as glyphosate. Understanding these mechanisms aids in the development of strategies to manage and mitigate weed resistance, such as altering herbicide application methods, implementing crop rotation practices, or exploring alternative weed control techniques in order to sustain an effective weed management system in agriculture.

A few soil microorganisms may utilize glyphosate as a source of nutrients, but under certain circumstances this may increase the population of pathogenic microbes. Glyphosate can alter the qualities of soil composition and inhibit the development of soil microbes. Investigations have revealed that both glyphosate and its metabolites have the potential to concentrate in soil and pollute aquatic environments, raising questions about the potential negative environmental impact that glyphosate possess. We emphasize the acute need of the hour or performing a comprehensive evaluation of exposure levels in nontarget organisms and risk evaluation of general population, which are pivotal to identify the serious side effects on human health.

■ ASSOCIATED CONTENT

Data Availability Statement

All data analyzed during this study are included within this article.

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Author Contributions

A.M. and T.M. designed the work. R.S. and G.K. have made significant contributions to conception and design, acquisition of data, analysis, and interpretation of data, as well as drafted the article. A.S. and M.G. drafted the article and made critical revisions. All authors have read and approved the final manuscript.

Notes

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■ ABBREVIATIONS:

ACCase: acetyl coenzyme A carboxylase
ADHD: attention deficit hyperactivity disorder
ALA: acetolactate synthase
AMPA: aminomethylphosphonic acid
ATSDR: Agency for Toxic Substances and Disease Registry
CAGR: compound annual growth rate
COVID-19: coronavirus disease 2019
2,4-D: 2,4-dichlorophenoxyacetic acid
DAHPS: 3-deoxy-D-arabino-heptulosonate 7-phosphate synthase
DNA: deoxyribonucleic acid
ECG: electrocardiogram
ECHA: European Chemical Agency
EFSA: European Food Safety Authority
EPA: Environmental Protection Agency
EPSP: 5-enolpyruvylshikimate-3-phosphate
EPSPS: 5-enolpyruvylshikimate 3-phosphate synthase
FAO: Food and Agriculture Organization
GBH: glyphosate-based herbicide
GE-HT: genetically engineered−herbicide tolerant
GM: genetically modified
GOX: glyphosate oxidoreductase
GR: glyphosate-resistant
GST: glutathione-*S*-transferase enzyme
HPPD: 4-hydroxyphenyl pyruvate dioxygenase
HR: herbicide resistant
IARC: International Agency for Research on Cancer
NHL: non-Hodgkin lymphoma
PEP: phosphoenolpyruvate
POEA: polyethoxyethyleneamide
PPE: personal protective equipment
RNAi: ribonucleic acid interference
S3P: shikimate-3-phosphate
USD: United states dollar
USDA: U.S Department of Agriculture
WHO: World Health Organization

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