

# Assessment of Polycyclic Aromatic Hydrocarbon (PAH) Pollution through Avian Sentinels: A Review of Blue Rock Pigeons

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**Abstract:** Polycyclic aromatic hydrocarbons (PAHs) are widespread environmental contaminants, known for their toxic and bioaccumulative effects on wildlife, particularly birds. These toxic compounds originate from anthropogenic and natural sources, leading to significant contamination risks. Various research methodologies, such as egg cell injection, yolk injection, eggshell application, and field studies involving controlled and uncontrolled groups at contaminated and reference sites, have been utilized to evaluate PAH contamination in avian species. However, existing studies predominantly focus on tracking short-term effects, often overlooking the long-term impacts and underlying mechanisms of PAH toxicity. Four to six-ring PAHs are particularly harmful, associated with adverse outcomes, including reduced body weight and fertility, enhanced organ development and metabolism, compromised immunity and survival rates, along with lower egg production and clutch size in birds. High incidences of reproductive disorders associated with PAHs have been documented in avian populations in the vicinity of human settlements or affected by oil spills in coastal and marine environments. Continuous exposure of PAHs is prevalent among bird populations. This review emphasizes the urgent need for a continuous monitoring program targeting PAHs and their metabolites across various bird species inhabiting both unaffected and polluted environments. Continued focus and further research into PAHs are crucial for understanding their long-term ecological impacts on avian health and biodiversity.

**Keywords:** Avian health, Bioaccumulation, Bioindicator species, Biomagnification, PAHs, Ecotoxicology, Reproductive disorders, Toxicokinetics, Wildlife pollution.

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## 1. Introduction

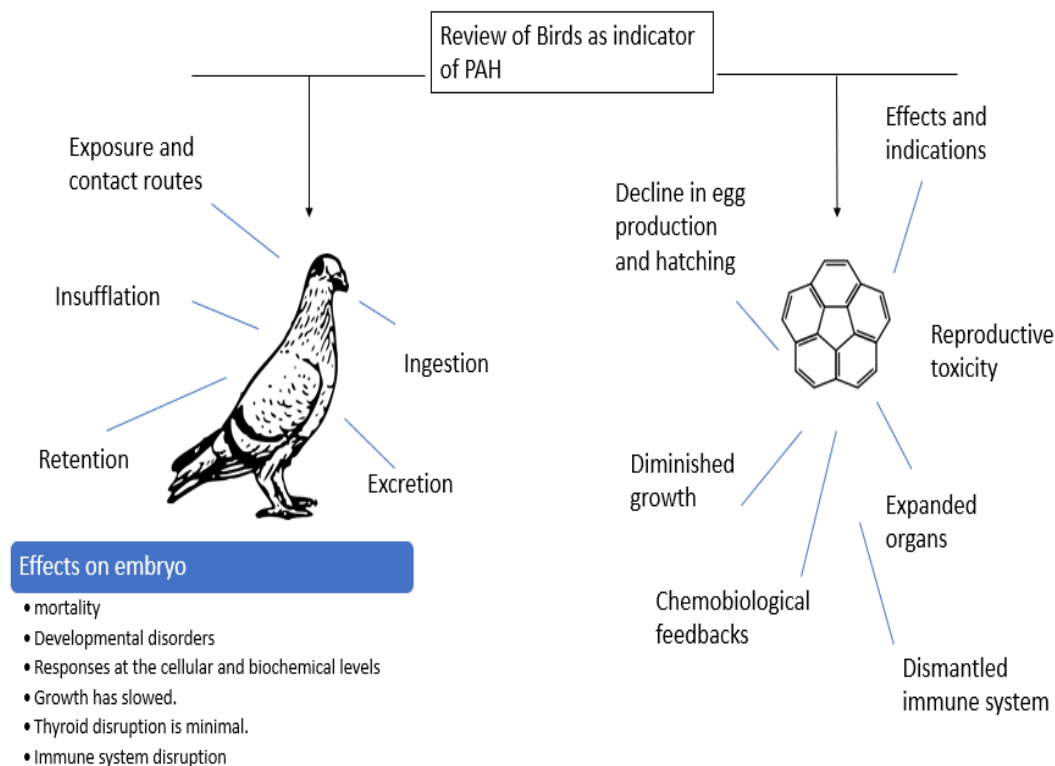
### 1.1. Polycyclic aromatic hydrocarbons

Polycyclic aromatic hydrocarbons (PAHs) are organic compounds consisting of two or more interconnected aromatic (benzene) rings composed exclusively of carbon and hydrogen (Mallah et al., 2022; Tartaglione et al., 2023). These compounds can transform into hydroxyl, quinoid, and nitrated metabolites (Ma and Wu, 2024; Manousi et al., 2021). PAHs are commonly found in petroleum, tar, and coal, as well as produced through incomplete combustion of organic matter. Coal combustion, automobile engines,

incinerators, biomass burning in forests, slash-and-burn agriculture, and smoking and cooking activities are among the major sources of PAH emissions (Chen et al., 2022; Wietzorek et al., 2022; Wang et al., 2022).

PAHs are colorless or pale-yellow solids, that can be found in air, water, soil, plants, and animal tissues around the globe. Due to their diverse structures and bioaccumulative properties PAHs exhibit varying degrees of toxicity and are persistent in the environment (Dey et al., 2023; Patel et al., 2020; Sahoo et al., 2020).

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## Graphical Abstract

Polycyclic aromatic hydrocarbons (PAHs) enter ecosystems through various natural and anthropogenic pathways. Natural sources include forest and brush fires, while anthropogenic sources encompass automobile emissions, cigarette smoke, and industrial activities. Interest in PAHs as a potential cause of illness surged in 1918 when the carcinogenic properties of benzo(a)pyrene were first identified (Anifowose et al., 2020; Arienzo et al., 2024; Famiyeh et al., 2021). Since then, extensive research has revealed the carcinogenic, mutagenic, teratogenic, and tumorigenic effects of PAHs, particularly in fish and mammals (Honda and Suzuki, 2020; Yebra-Pimentel et al., 2015). This growing body of evidence highlights the urgent need to monitor PAH exposure and develop strategies to mitigate their harmful effects on ecosystems and public health (Ephraim-Emmanuel and Ordinioha, 2021; Sekar and Praveenkumar, 2024). Understanding the sources, behavior, and environmental fate of PAHs is essential for assessing their ecological impact and the risks they pose to both human health and wildlife.

### 1.2. Structure and Formation of PAH

PAHs are mainly formed through incomplete combustion of organic materials. This process also contributes to the generation of combustion-derived

particles. Beyond their environmental impact, PAHs have significant roles in astrochemistry and materials science. These compounds are known carcinogens and persistent organic pollutants (Reizer et al., 2022).

A study investigates benzo(a)pyrene formation from chrysene and Benz (a)anthracene using a methyl addition/cyclization (MAC) mechanism. The study provides mechanistic insights into the chemical steps involved, including addition reactions, ring closures, hydrogen abstractions, and intramolecular hydrogen shifts. Results reveal that both reaction routes exhibit closely matched energetic trends, with an average energy difference of only 6.13 kJ/mol, highlighting the stability of the benzo(a)anthracene pathway (Almeida et al., 2012).

Heteroatoms and substituents on the ring system are absent from PAHs. Light PAHs are defined as those with less than four rings, and heavy PAHs are those with more rings (Eom et al., 2007). Heavy PAHs are more hazardous and stable than light PAHs. (Tkachenko et al., 2021).

PAHs are hydrocarbons ranging from naphthalene to coronene, i.e., two rings to seven rings, and chemically comprise two or more benzene rings fused into liner, cluster, or angular patterns with alternating single and double bonds. Phenanthrene and anthracene

are the simplest PAHs due to their smaller structures, i.e., they consist of only 3 rings (Bartonitz et al., 2020). PAHs have been classified into small (with up to six fused aromatic rings) and large (with more than seven fused aromatic rings) PAHs. Benzo(a)pyrene and 7,12-dimethyl benzo anthracene are the most extensively analyzed and studied PAHs.

Sometimes true PAHs are considered by investigators with nitrogen, sulphur, or oxygen within the ring or substituted for bonded hydrogen. Most PAHs are formed by pyrolysis and subsequent synthesis of the organic matter (Pereira, 2014).

In the presence of high temperatures, i.e., 500-800 degrees Celsius, PAHs are formed because of incomplete combustion of organic matter, whereas in low temperatures, i.e., 100-300 degrees Celsius, by subjecting organic matter to sediments, PAHs production is observed in the form of coal and oil in

sedimentary rocks, over a long period. (Zhang et al., 2015).

Although PAHs formed in both temperatures are identical, alkylated compounds formed at low temperatures are abundant.

### 1.3. Fundamental toxic nature

Cancer is the foremost genuine result of PAH harming. PAHs have a mooo intense harmfulness. Based on PAH, a few considers have found noncarcinogenic impacts. After drawn out introduction, the non-carcinogenic impacts of PAHs to a great extent influence the pneumonic, gastrointestinal, renal, and dermatologic frameworks. Numerous PAHs are either humbly mutagenic or non-mutagenic in vitro; in any case, their metabolites or subsidiaries can be solid mutagens (Kobetičová et al., 2008).

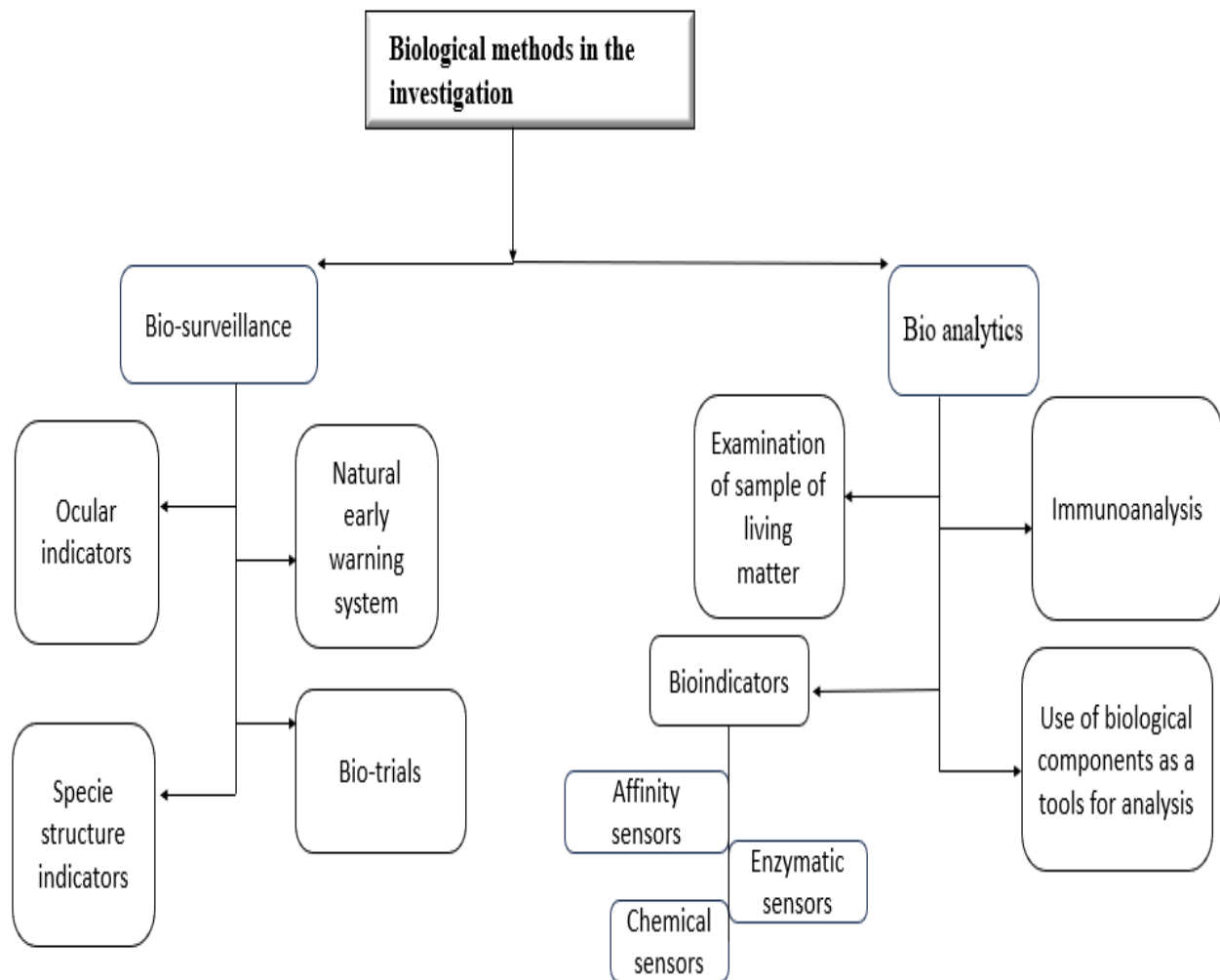


Fig. 2. Biological Methods for Analyzing Environmental Pollutants, Focusing on Polycyclic Aromatic Hydrocarbons (PAHs).

#### 1.4. Routes of exposure for polycyclic aromatic compounds

Most people endure exposure to PAHs daily through the air, water, soil, and food. Exposure modalities include intake, inhalation, and skin contact in both occupational and non-occupational circumstances (Pereira et al., 2009). Various exposures, such as cutaneous and inhalation exposures from polluted air, may include numerous routes at the same time, which impacts the total absorbed dose. All non-workplace sources of exposure should be evaluated, including food, smoking, and coal and wood combustion (Dhananjayan, 2013).

#### 1.5. Bioaccumulation of PAH in living organisms

PAHs have the ability to be absorbed and stored by living beings due to their lipophilicity. Several investigations have demonstrated that PAHs can accumulate in rodents, pigs, cows, and human adipose tissue (Luzardo et al., 2014).

Immunotoxicity, embryonic abnormalities, and cardiotoxicity in aquatic life. Tumours, reproduction, development, and immunity in birds. Skin, lung, bladder, liver, and stomach cancers are more common in animals, as are injection-site sarcomas. Skin, lung, bladder, and gastrointestinal malignancies are more common in humans (Power et al., 2021).

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## 2. Significance of birds concerning ecology and environmental pollution

Birds are a big and diverse group of species that play many important ecological roles and offer a multitude of ecosystem services. As intermediaries between apex predators and nutrients, they actively participate in the trophic transfer of pollutants (Brausch et al., 2010).

Birds maintain plant diversity and growth and also perform important ecological roles as pollinators and seed dispersers. Several birds can function as ecosystem engineers in some habitats (e.g., Little Auk, Black-backed Woodpeckers, and vultures of the Accipitridae and Bucerotidae families) or are keystone species. Birds have cultural significance and have represented countries and groups (Franci et al., 2018).

Sadly, reports of avian population decrease and extinctions in North America and other parts of the world have been linked to habitat degradation, unrestricted harvesting, and human causes of mortality. When it comes to chemical modeling, risk assessment, environmental benchmarking, and the management of chemical pollutants, birds are a significant class of vertebrates to take into account. In the past, it has been

documented those birds are among the species that experience negative effects from pollution. (Ball and Truskewycz, 2013).

During the 1850s industrial revolution, reports of manmade environmental pollutants harming birds and other species started to mount. These reports included cases of hydrogen sulfide fumes near oil fields, industrial smokestack emission toxicity, and arsenic and lead poisoning. Birds have long been utilized as sentinel species because of their great chemical sensitivity, which makes them effective bioindicators of environmental contamination (Ball and Truskewycz, 2013).

Fowls are in a perfect world suited to comprehend and evaluate the ultimate destiny and ecotoxicological results of natural compounds in differing settings since they are plenteous apex predators in a assortment of environments. They can help within the progression of comparable investigation in other vertebrates. Because of their tall trophic position, long life expectancy, and huge scavenging territories that permit for spatiotemporal introduction and ecotoxicological destiny evaluations of pertinent chemicals, winged creatures of prey are frequently utilized as sentinels for natural contamination. Fowls have generally been uncovered to a wide run of toxins. One of the foremost well-known cases is the enormous worldwide decrease in raptor populaces caused by eggshell diminishing after presentation to dichloro-diphenyl-trichloroethane (DDT). Intense inebriation from poisons regularly appears like a less strong and quick danger, but extreme indirect toxic effects, basically agrochemicals, on farmland fowls are depicted as drivers of population decline (Bartonitz et al., 2020).

Other threats at the population level associated with chemical exposure have been reported for avian taxa, such as global lead poisonings of raptor and waterbird species. Nevertheless, as of right now, no comprehensive long-term survey has been conducted on birds, either individually or in populations, to evaluate the short- or long-term impacts of the several chemicals or chemical mixes.

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## 3. Exposure of Birds to PAH

Birds pose unique and significant challenges to study bioaccumulation and biomagnification of pollutants. Unlike fish and other aquatic invertebrates, which are often confined to specific and localized habitats, birds show remarkable mobility and are widely associated with all major natural spheres where life can exist (e.g., atmosphere, lithosphere and hydrosphere). This wide range of habitat makes it

difficult to ascertain the conditions under which they are exposed to various chemicals.

The assessment of bioaccumulation of contaminant in birds is complicated by several factors, including their diverse diets and the dynamic nature of their habitats. Environmental changes, such as shifts in natural niches and climate change, further complicate the evaluation of contaminant levels within avian populations. Moreover, different bird species vary in their suitability for chemical monitoring based on ecological traits such as migratory behavior and feeding environments (Head et al., 2015).

Bioaccumulation and biomagnification are regulated by a complex interplay of chemical properties of pollutants, distribution sources, trophic interactions, and species biology. Interspecies differences in gut physiology, dietary preferences, foraging strategies, ecological mobility, physiological characteristics, and other species-specific traits significantly affect uptake and metabolism of environmental contaminants by birds (Alonso-Alvarez et al., 2007). These factors will be briefly reviewed in the following sections.

Contaminants have been associated with the decline in bird population and chronic sublethal effects. For example, oil spills have resulted in long-term consequences such as reduced reproductive success and altered demographics due to various environmental stresses, including decreased food availability. High-profile cases of contamination affecting birds include lead poisoning in the White-tailed Hawk and California Condor (*Gymnogyps californianus*), the decline of Gyps vulture populations due to diclofenac exposure, and pesticide-related poisoning in the Ruddy Kite (*Milvus milvus*).

Recent reviews have highlighted chemically contaminated diets as a significant anthropogenic threat to avian scavenger species. These findings supported the well-documented declines in populations of the Peregrine Falcon, Sparrowhawk (*Accipiter nisus*), and Bare-faced Ibis following the widespread application of DDT over 60 years ago. This revision enhances clarity, coherence, and engagement while maintaining the original content's integrity.

### 3.1. Biotransformation of Birds

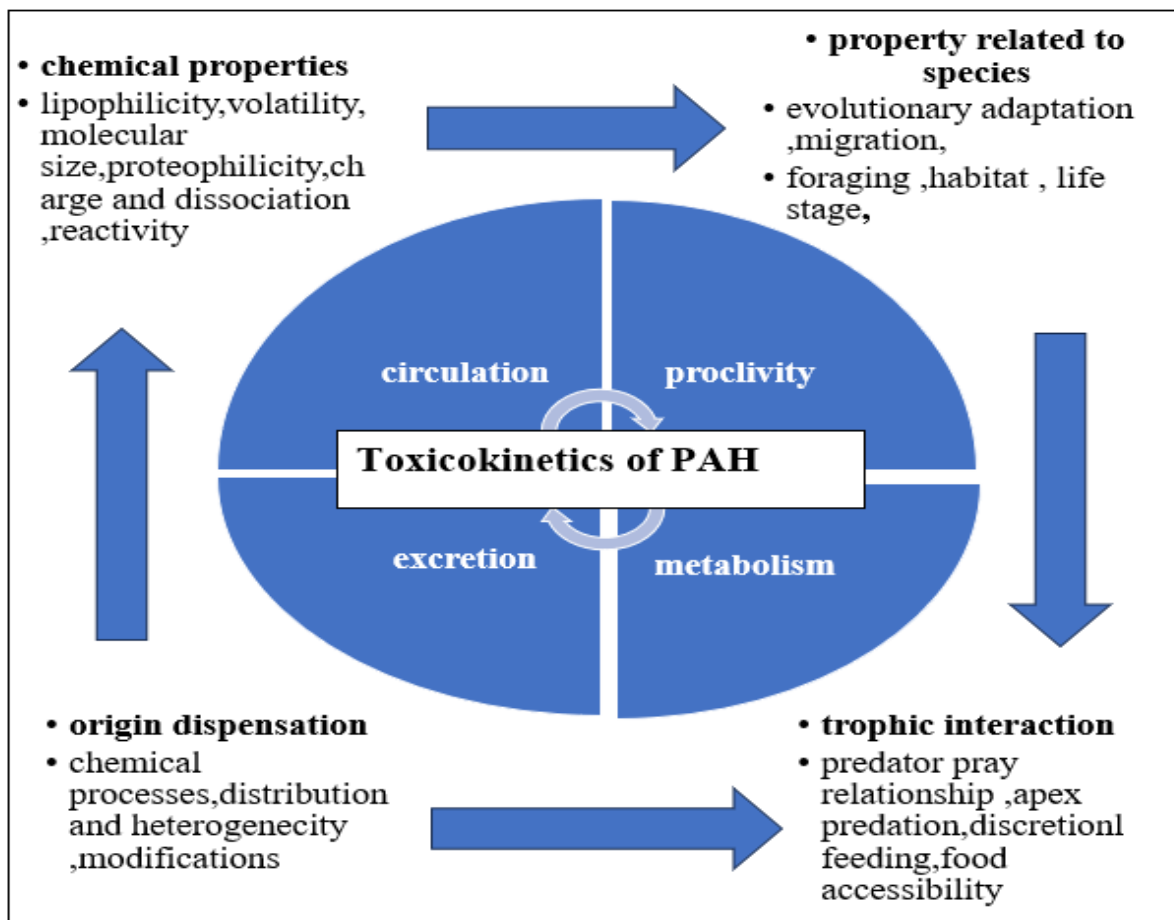
One of the most critical processes for understanding the uptake, elimination, biological toxicology, fate, and bioaccumulation of organic chemicals in exposed organisms, such as birds, is biotransformation (Fig. 2) (Kuo et al., 2022; Zhang et al., 2021). The importance of biotransformation in ecotoxicology has been recognized since early studies on fish, worms, and other invertebrates (Wang et al., 2024; Xue et al., 2023). In fact, biotransformation and chemical partitioning play a more significant role than growth dilution, fecal elimination, or even bioavailability correction for dissolved or particulate organic matter in determining reliable estimates of the bioconcentration factor of organic compounds in fish (Burkhard, 2021; Gómez-Regalado et al., 2023; Teunen et al., 2021).

### 3.2. Effects on Birds (especially blue-rock pigeon)

Birds can suffer from a range of adverse effects due to exposure to PAHs, including developmental defects, reproductive issues, respiratory problems, altered behaviors, immunosuppression, bioaccumulation, and genetic damage. These contaminants not only disrupt ecosystems but also contribute to population declines in avian species. To safeguard bird populations and their habitats, effective mitigation measures are essential. These include reducing PAH release, improving waste management practices, and monitoring polluted areas.

A study investigating the toxicity of PAHs determined the LD50 values for six PAH congeners in blue rock pigeon and quail embryos, highlighting their vulnerability to PAH exposure. The results indicated dose-dependent embryo mortality with limited additional effects. Notably, the sensitivity observed in both chicken and quail embryos enhances our understanding of PAH toxicity across avian species.

Another study focused on native and alkylated PAHs in greater scaup and great crested grebe tissues. PAH distribution varied across bird tissues. Maximum concentrations were recorded in scaup lungs and grebe kidneys. Alkylated PAHs accounted for a sizable part of the total PAH composition. These variations in PAH profiles were most likely caused by metabolic differences rather than dietary differences.



**Fig. 3. Chemical Properties and Factors Influencing Distribution and Toxicokinetics**

Importantly, the PAH levels discovered in these birds were relatively modest when compared to global data, indicating that they posed no substantial health risk.

In examining the impacts of PAHs on blue rock pigeon embryos, various forms of PAHs—including individual compounds, mixtures, refined products, and crude petroleum—were utilized. This approach was adopted because rapid detrimental effects from oil pollution can be observed in birds

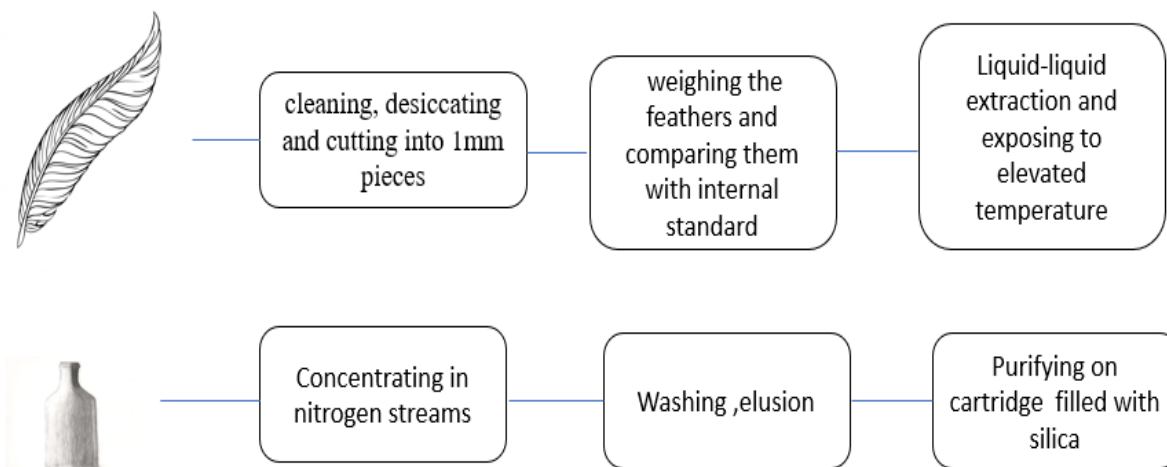
Different methods were employed for exposure: PAHs were injected into egg yolk or air spaces, applied to eggshells, or immersed for periods ranging from three to ten days for exposure and three days to two weeks for reaction. The most commonly used bird species for yolk applications were blue rock pigeons, while mallards were primarily used for eggshell applications; turkeys and common ducks were also included in some studies.

PAHs can be hazardous to birds in the short term and have long-term consequences. Among these effects are given below.

Egg production and hatching have been reduced. Increased brood abandonment or clutch abandonment. Growth has slowed. Weight gain in the organs. Several biochemical responses. PAHs can also be harmful to reproduction, development, and immunity. The PAH Benzo(a)pyrene is a well-known carcinogen and mutagen. PAHs are emitted when certain organic compounds are incompletely burned.

### 3.3. Presence and impacts of PAH in the egg yolk of blue rock pigeon

A study investigated the transfer kinetics of Persistent Organic Pollutants (POPs), specifically PAHs and lindane, into egg yolks of Hyline hens. The hens were administered these contaminants, and the results showed that PAHs, mainly as metabolites, entered yolks rapidly with low overall recovery rates.



**Fig. 4. Liquid-Liquid Extraction Method for Analyzing Environmental Pollution.**

In contrast, lindane had slower elimination via eggs, likely due to body fat storage. These findings highlight the mechanism by which persistent organic pollutants (POPs) are transferred to and metabolized within egg yolk.

Studies reported that introduction of relatively low doses (2.2-20ng/egg) of highly toxic PAHs including benzo(k)fluoranthene, induced significant cellular damage, biochemical responses, and reduced embryonic survival (Albers and Loughlin, 2003). Contrarily, higher doses (100-600ng/egg) administered via yolk injection produce comparatively less severe effects.

This greater sensitivity associated with air cell exposure can be attributed to rapid diffusion of PAHs into the highly vascularized chorioallantois membranes, which develops extensively between day 3 and 10 of incubation (Wallace et al., 2020). Conversely, lipophilic nature of PAHs leads to slower transfer when injection to lipid-rich yolk, delaying embryonic exposure.

Both exposure routes are useful for assessing PAH toxicity. Chicken eggs showed a greater decrease in survival and liver necrosis when coal tar fractions of PAH containing five to six rings were injected into the egg yolk. PAH fraction of air particulate extract injection into the air space of the egg caused a decreased survival rate of the embryo and this was the case in four to seven-ringed PAH. Halogenated hydrocarbons are the most potent source of microsomal P450-associated monooxygenase activity as compared to polycyclic aromatic hydrocarbons. The reason over here is the greatest metabolism of PAH in short-term exposure in the case of vertebrates i.e., birds

and toxicity of PAH is strongly dependent upon aryl hydrocarbon (AH) receptor and AH binding resulting in interruption in cellular membrane function, enzyme system, and dysregulation in gene expression upon exposure to PAH (Honda and Suzuki 2020).

### 3.4. Embryos of birds especially blue rock pigeons: influenced by PAH toxicity

The study focused on the maternal transfer of PAHs in the blue rock pigeon. Researchers analyzed PAH levels in maternal liver and uterine content (eggs and early-stage embryos) to determine the transfer rate to offspring (Dhananjayan, 2013). They found a mean transfer rate of 13%, with significant variability among individual PAH congeners, with benzo[b]fluoranthene having the highest transfer rates. The transfer rates were influenced by the physicochemical properties of each PAH, with lower molecular weight PAHs showing higher transfer rates. The study did not find clear evidence of a depuration mechanism related to female size or consecutive reproductive cycles. Overall, the research highlights the potential exposure of endangered embryos to PAHs, raising concerns about possible harmful effects on their development and emphasizing the need for conservation efforts (Goodchild et al., 2020).

Four to six-ring compounds pose serious toxicity to bird embryos. The PAH compounds are responsible for the highest microsomal, P450-associated monooxygenase activity in fishes. 16 PAH observed in four sample species of seabird eggs were different in individual and total concentration i.e., nondetectable to 6.77ng/g and 1.59ng/g to 8.44ng/g respectively (Pereira et al., 2008). These concentrations were in the lower limit of air cell injection by effect threshold,

threefold the effect threshold in eggshell application, and less than the effect threshold for yolk injection.

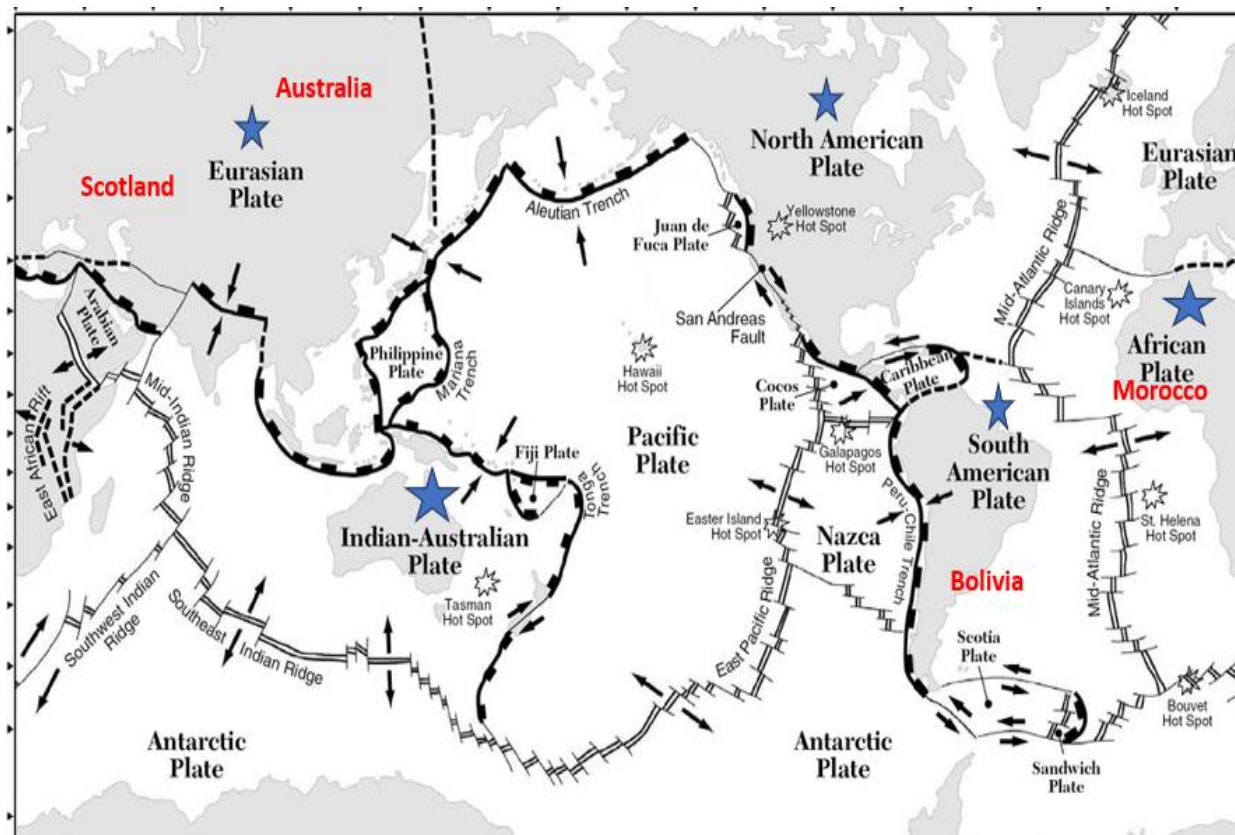
In comparison, nine eggs of abandoned peregrine falcons collected after the prestige oil spill on the coast of Spain showed 295ng/g and 21ng/g to 461ng/g for individual and total PAH concentrations respectively. Concentrations were within or above the effect threshold for air cell injection and among these three eggs were within the effect threshold for egg yolk injection. Greater loss in clutch size was observed when compared to pre-spill size affecting physiological response and survival of the embryo (Pereira et al., 2008).

### 3.5. PAH and Birds- Case Studies and Symptoms

In the late 1970s and 1980's effects of crude oil were studied on herring gulls and retarded weight gain, increased organ weight and increased endocrinal system activity was observed by administration of a single dose of 0.1 to 1ml and an occasion of five daily doses of 10ml/kg to chicks (Albers and Loughlin, 2003). An increased hepatic mixed function activity

and liver enlargement were observed in the case of administration of aromatic fraction. Mallard ducklings kept in an oil sand wastewater storage for thirty-three days, suffered from reduced weight gain, skeletal structure problems, and increased liver metabolites of pyrene and naphthalene in contrast to referenced water storage site (Albers and Loughlin, 2003)

Measurement of chemical detection or biochemical response observation is done to detect PAH toxicity in uncontrolled adult birds. It is difficult to tell the number and total PAH detected due to variation in analysis so this is suitable for intra study. However, for both inter as well as intra-study quantification and biochemical responses to PAH are helpful (Pei et al., 2017). The illustrations are: 0.01 to 0.59 mg/kg concentrations of individual PAH are found in skin, fat, and carcasses of double-crested cormorants and wintering waterfowls. Wintering adults in freshwater industrial areas has 0.07mg/kg concentration of individual PAH resulting in chromosomal damage (Fernie et al., 2018).



**Fig. 5. Distribution of Blue Rock Pigeon Populations and Sampling Locations.** This figure illustrates the sampling regions, highlighting areas inhabited by Blue Rock Pigeons. The countries marked in red indicate significant populations of Blue Rock Pigeons, while the blue star denotes the specific regions where our studies were conducted. **Source:** Originally produced by the author.

Near a refinery site on North Platte River, Casper, Wyoming, USA, and at a reference site i.e., 10km upstream eggs and chicks of tree swallow and house wren were taken and it was observed that PAH concentration at the refinery site in both was much higher as compared to reference site and the results were consistent with concentrations in sediments and chick dietary samples. It was observed that methylated PAH was not in much concentration in sediment, diet, and bird carcasses showing that PAH from petroleum was not the culprit but the PAH from the combustion was. Less than 0.25 ratio of pristine as well as phytane and higher odd-numbered aliphatic hydrocarbons in chick and diet samples showed that wren and swallow chicks were not chronically exposed to petroleum (Dhananjayan, 2013). In the livers and eggs of swallows and wrens, trace concentrations at the reference site were the same or more than refinery site and concentrations of strontium, selenium, and boron were high in eggs and livers of swallows and wrens at both sites (Fernie et al., 2018).

Gannet eggs, from Ailsa Craig and Bass rock i.e., two Scottish colonies, of golden eagles from Hebrides and highlands, and of merlin eggs from Scottish borders were taken and analyzed for 52 PAHs i.e., two to seven-ringed parent and alkylated polycyclic aromatic hydrocarbon compounds. Phenanthrene from Ailsa Craig was in abundance and methylanthrene was in abundance in other locations and in other species. No embryotoxic concentrations were detected, although in eggs, PAHs were detected. 3.1 to 5.7 ng/g wet wt. of PAH was analyzed but this was a uniform and low-level accumulation suggesting background exposure to diffused sources.

Three-ringed parent and alkylated PAH were detected in eggs of marlins and gannets from Ailsa Craig and Hebridean golden eagles, whereas mixed profiles were present in other eggs (Wallace et al., 2020).

#### 4. Conclusion

Due to lipophilic, bioaccumulative and persistent nature of PAH, they are disease producing contaminants causing reproductive disorders, immunity deficiencies, hormonal problems and several others in birds. Although PAHs are metabolized to great extent in birds, due to their higher metabolisms yet parent concentrations are high enough to show symptoms. In different case studies, it was found that ducks are more susceptible to PAH as compared to domesticated chicken. Enhanced metabolic activity,

growth abnormalities and cellular damages are the results of PAH injections. However, embryos are highly affected by air cell injections of four to six ringed PAHs. Young birds affected by oil spills showed increased organ growth, reduced weights and several endocrinal disorders along with immunity problems. Reduced egg production, decreased survival rates and liver dysfunctions and damages were observed in adult birds. The exact mechanism of PAH contamination is not known in birds however through enzyme systems, genes expressions some of the tracking studies have been done. Birds living near oil spill areas are affected by immediate and chronic effects like deaths and decreased fertility problems respectively. Blue rock pigeons can be used as a bioindicator specie for the continuous monitoring of PAH in a residential or urban area. PAH level can be analyzed through examination of the liver in case of blue rock pigeon as well as it can be used as a tool of checking recent exposure of PAH in environment and the continuous input in urban environment. Exxon Valdez oil spill is indicator of immediate impacts on birds and shows short term effects but long-term effects of PAH mechanisms are still not much clear. Although recently on vultures in India, a study has done, on tissues of birds showing bioaccumulative effects of PAH leading to reproductive problems in birds i.e., decreased bird count in last ten years. Oil spills are the greatest source of PAH contamination specially in sea birds by their external oiling or by ingestion.

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