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A Review On Impact Of Birth Control Pills In Muncipal Wastewater On Male Fish Population

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Nisha Rana*, Manu Varma and Seema Jain*****

Abstract

The presence of estrogenic compounds (natural estrogens and synthetic chemicals that mimic natural estrogen) in municipal wastewater potential harm to aquatic life and also affect the reproductive health of wild fishes. Frequently, one particular synthetic estrogen has been singled out for purportedly detrimental effects on the environment: ethynyl-estradiol, or EE2, a synthetic estrogen used in birth control pills, patches, rings, and injectables. Synthetic estrogen in the women's urine goes through sewage treatment plants without being completely broken down, and the fish absorbs it, with negative impact. Male fish produce eggs in their testes. Female fish are stimulated by the extra hormones to produce eggs at the wrong times of year. Male fishes found in downstream of some wastewater outfalls produce vitellogenin (VTG) and early-stage eggs in their testes, and this feminization has been attributed to the presence of estrogenic substances such as natural estrogens[estrone or 17β-estradiol (E2)], the synthetic estrogen used in birth-control pills [17α-ethynyl estradiol (EE2)], or weak estrogen mimics such as nonylphenol in the water. Earlier research suggested that fishes inhabiting waters that receive untreated municipal wastewaters or effluents from municipal wastewater treatment plants (MWTPs) are exposed to chemicals that affect reproductive endocrine function and also the "gender-bending" effect in fish downstream from sewage plants. The present review suggests that the concentrations of estrogens and their mimics observed in freshwaters can impact the sustainability of wild fish populations. Lately these compounds have become a major concern since they influence sexual differentiation and reproductive success of male fishes.

Keywords: *Municipal Wastewaters, Ethynyl-Estradiol, Estrogen, Male Fishes, Endocrine function.*

&&***Dept. of Z.oology, R.G. P.G. College, Meerut*

Introduction

Estrogenic compounds are a class of pharmaceutical products harmful to the animals and a cause of environmental damage. The biological activity of these compounds is high, which are designed to operate at low concentrations. Therefore, even in low concentrations in the environment can have harmful effects on aquatic organisms and in humans, who might be consuming water or food contaminated with estrogen. Synthetic estrogen in the women's urine goes through sewage treatment plants without being completely broken down, and the fish absorb it, with bad effects following. Birth control pills are not totally absorbed in the body and can reach waterways by being flushed down toilets, poured down sinks and excreted in urine. The estrogenic chemical can gather in fairly heavy doses. 17á-ethinylestradiol has been known to cause adverse reproductive effects including reduced fecundity and fertility, intersex and sex change in fish by mimicking naturally produced estrogen at low concentrations (Armstrong, 2011). Endocrine-active substances (EASs) have been shown to affect several species of fish in different parts of the world. In particular, feminizing effects have been observed in male fish. Attempts to better understand causal factors affecting estrogenicity in municipal wastewater have primarily focused on analytical evaluation of specific chemical estrogens and the use of estrogen receptor (ER) based *in vitro* assays (Huggett,2003). The present review work was conducted on impact of birth control pills in municipal wastewater on male fish population. The hormone, called ethyny lestradiol or EE2, is one of many chemicals that can disrupt the endocrine system, which regulates animals' growth and development. U.S.G.S. (2008) reported that male fish became "feminized" when exposed to human hormones. Some of the fish, a type of fathead minnow, produced early-stage eggs in their testes while others actually developed tissues for both reproductive organs. It seems that widespread use of birth control pills has elevated the amount of estrogenic substances going into our waste stream; researchers actually added the synthetic estrogen found in contraceptive pills to a remote lake in northern Ontario in amounts that are normally found in human wastewater (Blenchfield, 2015). Despite widespread recognition that municipal wastewaters contain natural and synthetic estrogens, which interfere with development and reproduction of fishes in freshwaters worldwide, there are limited data on the extent to which natural populations of fish can recover from exposure to these compounds. Whole-lake additions of an active component of the birth control pill

(17á-ethynylestradiol; EE2) that resulted in the collapse of the fathead minnow (*Pimephales promelas*) population (Blanchfield, 2015). The male fish developed some feminized characteristics, such as producing proteins normally synthesized in females. But what really disturbed the scientists was how populations of the fish crashed to near extinction levels by the end of the experiment. Feminization of the males combined with hormonal changes to the females apparently damaged their overall reproductive capacity to the point that the fish were unable to maintain their population. The continued inputs of natural and synthetic estrogens and estrogen mimics to the aquatic environment in municipal wastewaters could decrease the reproductive success and sustainability of fish populations. Knowledge of possible toxic mechanisms (or modes) of action (MOA) of chemicals can provide valuable insights as to appropriate methods for assessing exposure and effects, thereby reducing uncertainties related to extrapolation across species, endpoints and chemical structure. However, MOA-based testing seldom has been used for assessing the ecological risk of chemicals (Gerald et al., 2009). The large-scale field study in the Netherlands, was conducted on the potential effects of estrogenic compounds on wild fish. The freshwater bream (*Abramis brama*) and the estuarine flounder (*Platichthys flesus*) were sampled at a large number of locations in the spring and fall of 1999 (Lahr *et. al.*, 2006).

Numerous studies have shown that estrogen and chemicals that behave like it have a feminizing effect on male fish and can alter female-to-male ratios (Bandari, 2008). Male fish produce eggs in their testes. Female fish are stimulated by the extra hormones to produce eggs at the wrong times of year. Scientists have seen this "gender-bending" effect in fish downstream from sewage plants, but lacked proof that birth control pills are a cause. Estrogen can wreak reproductive havoc on some fish, which spawn infertile offspring sporting a mixture of male and female parts. Studies of fish upstream and downstream of wastewater treatment plants have found more female and intersex fish downstream from the plants, presumably because of the higher estrogen levels in the downstream water. It is recommended that the 30-d average concentration of 17á-ethinylestradiol (EE2) in water should not exceed 0.5 ng L-1 with no single value to exceed 0.75 ng L-1 (no more than 50% above the guideline value) to protect freshwater aquatic life from adverse effects (Nagpal and Meays 2009).

EE2: Source And Effect On Fish

EE2 [17-ethynyl estradiol (EE2)], a synthetic hormone, is only one of a cocktail of natural and synthetic hormones that humans excrete into wastewater, including other estrogens. Sources of estrogen include birth control pills and postmenopausal hormone treatments, as well as the estrogen that women produce naturally and excrete Gustavo (2014). EE2 is a major ingredient in oral contraceptives for women, and up to 68 percent of each dose is released in the latrine through urine and excrement. A full dose is released when some women simply pour unused pills down the drain. EE2 is endocrine disruptor that interfere with hormones and cause developmental disorders. The generic term (estrogen) applies to any substance, natural or synthetic exert biological effects characteristic of ovarian estrogenic hormones, many anabolic and growth promoters in animals, which has led to its misuse in the commercial animal. Most pharmaceutical estrogens and xenoestrogens are introduced into the environment through municipal waste water treatment plant (WWTP) effluent sources. Less than 1 ng/L EE2 can cause feminization of male fishes, 4 ng/L caused abnormal reproductive development (male fathead minnows). E2 has been detected at concentrations from 1 ng/L to 80 ng/L. Maxine et. al., 2015 recorded Nonylphenol, a surfactant and brominated biphenyls, a flame retardant have been detected between 0.1-3.7 ìg/L and 0.3-4.6 mg/kg (on suspended particles) respectively. Total estrogenicity (E2 equivalents) of 147 ng/L has been measured in WWTP effluent. The Gender-bending is happening at several locations on the few Rivers, showing up in between 50 and 100 percent of bass caught and dissected. The body of a fish reacts to EE2 as if it were a natural estrogen, "demasculinizing" male animals and creating a condition called intersex that interferes with an animal's ability to reproduce. There is considerable evidence that fishes inhabiting waters that receive untreated municipal waste waters or effluents from municipal wastewater treatment plants (MWTPs) are exposed to chemicals that affect reproductive endocrine function. Male fish downstream of some wastewater outfalls produce vitellogenin (VTG) mRNA and protein, associated with oocyte maturation in females, and early-stage eggs in their testes. This feminization has been linked to the presence of estrogenic substance ssuch as the natural estrogen 17-estradiol (E2) and the synthetic estrogen 17-ethynylestradiol (EE2) (Walter et al., 2015).

Intersex males often produce eggs in their testes. After the active

ingredient in most birth control pills has done its duty preventing pregnancy, it begins a second life as a pollutant that can harm wildlife in waterways. Environmental estrogens in the aquatic environment have been shown to be responsible for the feminization of fish. The estrogenic content of the Yangtze River (Nanjing section) was assessed using a combination of bioassay and chemical analysis. The in vivo bioassay was conducted by exposing adult male goldfish (*Carassius auratus*) to different concentrations of river water (25%, 50% and 100%) sampled from three representative sections of the studied area. The results of these in vivo bioassay and chemical analysis demonstrate that fish in the Yangtze River are exposed to environmental estrogens and are at a risk of feminization (Lu *et. al.*, 2010).

Treatment of EE2

Wastewater treatment facilities have become necessary in ensuring the discharges of high quality wastewater effluents into receiving water bodies and as consequence, a healthier environment (Amenu, 2014). The European Union is the first entity to seriously consider mandating the removal of ethinyl estradiol, also known as EE2, from wastewater. The treatment of the water is not possible as lack of proper management. EE2 remains even after wastewater is treated, so it's a safe bet that large doses arrive when sewage facilities dump millions of gallons of *untreated* wastewater into the Potomac and other waterways during overflow events when pipes are overwhelmed by rain (USGS, 2015). The charcoal based method can be used for removing EE2. Charcoal-based filtering is the most effective method for removing chemicals such as EE2 from wastewater. Other methods may also be effective, but they, too, these are very expensive to apply, and require some financial investment. It will cost quite a lot of money to upgrade to meet environmental quality standards.

Regardless of the source of the contaminated biomass, when the land sits above a shallow drinking water sources, estrogenic compounds can enter into groundwater. In rural areas with less comprehensive sewage treatment, the risk of groundwater contamination may also increase. Scientists for King County's Department of Natural Resources and Parks, which conducted the recent survey, suggested that a synthetic female hormone used in birth-control pills and hormone-replacement therapy has been found in streams and lakes, upstream from any sewage-treatment plants. That suggests that the prescription drugs are getting into the water from septic tanks or leaking sewer pipes. One-third of male Pearl Dace minnows grew eggs in their testes. The

entire population of the common Fathead minnow, once numbering in the several thousands, crashed to near zero because the hormone-stoked fish couldn't reproduce. Natural and synthetic estrogens are not completely broken down in current MWTP processes, and, as a result, are discharged into receiving waters in both treated and untreated wastewaters and found in the aquatic environment at low parts per trillion concentrations (typically<5 $ng \cdot L^{-1}$;). These effluents contain mixtures of individual estrogens and their mimics that differ in their ability to elicit estrogenic responses. For this reason, total estrogenicity (expressed as E2 equivalents) of an effluent or water sample is determined either by summing concentrations of individual compounds after adjusting these concentrations by the compound's estrogenic potency (relative to E2) or with bioassays. Using these approaches, total E2 equivalents of up to 147 and 17 $ng \cdot L^{-1}$ have been measured in final effluents and surface waters, respectively. Within this group of substances, the estrogen used in birth-control pills, EE2, is one of the more potent estrogens present and has been linked to the feminization of male fishes in river receiving municipal wastewater.

Earlier Evidence

At the Experimental Lakes Area in northwestern Ontario, Canada, a long-term, whole-lake experiment was conducted using a before-after-control-impact design to determine both direct and indirect effects of the synthetic oestrogen used in the birth control pill, 17a-ethynyloestradiol (EE2)(Kidd et. al., 2007). Experimental Lakes Area (ELA) in north-western Ontario, Canada, to assess the subcellular-level through population-level effects of the potent synthetic estrogen EE2 on fathead minnow. The concentrations of EE2 achieved in the experimental lake, Lake 260, during the 3 years of additions were within the range of those observed in untreated and treated municipal wastewaters (Palace *et. al.*, 2006) and below the total E2 equivalents (with EE2 having an E2 equivalent of 0.19–1.9) measured in effluents and receiving waters. In the fish tested for the study, a 30 percent drop in fertilization rates was observed. While the exposed fish and their immediate offspring appeared unaffected, the second generation of fish struggled to fertilize eggs — with a 30% reduction in fertilization rates — and their embryos were less likely to survive. Even the third generation of fish had 20% impaired fertility and survival rates, though they were never directly exposed to the hormone. At the Experimental Lakes Area in northwestern Ontario, Canada, a long-term, whole-lake experiment was conducted using a before-after-control-impact design to determine both direct and indirect effects

of the synthetic oestrogen used in the birth control pill, 17a-ethynyloestradiol (EE2). Algal, microbial, zooplankton and benthic invertebrate communities showed no declines in abundance during three summers of EE2 additions (5–6 ng l21), indicating no direct toxic effects. Recruitment of fathead minnow (Pimephales promelas) failed, leading to a near-extirpation of this species both 2 years during (young-of-year, YOY) and 2 years following (adults and YOY) EE2 additions. Body condition of male lake trout (Salvelinus namaycush) and male and female white sucker (Catostomus commersonii) declined before changes in prey abundance, suggesting direct effects of EE2 on this endpoint. Evidence of indirect effects of EE2 was also observed (Kidd, 2015). Potential effects of exposure to the synthetic estrogen 17a-ethynylestradiol (EE2) were examined in several species of fish from a lake experimentally treated with environmentally relevant concentrations of the contaminant (Palace et. al., 2005). Campbell (2006) suggested that due to their persistent and ubiquitous nature, source control strategies for e-EDCs may reduce influent concentration to wastewater treatment plants so that the post treatment effluent will decrease concentrations to estrogenically inactive levels. Alternatively if source reduction is not possible, then more testing is needed on tertiary treatment technologies and treatment efficiencies for e-EDCs. There is still a need for research on remediation and restoration approaches for habitats disturbed by elevated e-EDC concentrations. EE2 induces biochemical and physiological effects in exposed fish, but linkages to widespread reproductive dysfunction in populations have not been established. Mortality in early life stages has only been documented at relatively high concentrations, above those found in the environment. To examine the potential effects of environmentally relevant concentrations of EE2, reproductive endpoints were examined in lake trout (*Salvelinus namaycush*) captured from a lake experimentally treated with ~5 ng/L EE2 (Werner, 2006). Disrupted population dynamics due to direct and transgenerational effects on survival and fecundity. Schwindt (2014) conducted a year-long study on three generations of fathead minnows *Pimephales promelas* Rafinesque in aquatic mesocosms and laboratory aquaria and added environmentally relevant concentrations of EE2 daily using a static renewal, which approximates a pulsed exposure that fish experience in natural systems (Schwindt, 2014). Evaluation of Estrogenic Activities of Aquatic Herbicides and Surfactants Using an Rainbow Trout Vitellogenin Assay was also studied by Xie et. al., in 2005. USGS scientists found that one-third of 111

American waterways they tested contained some intersex fish, particularly male bass. A year later, scientists were reporting that 80% of the fish in the Potomac River — whose water is pumped into the homes of 4 million people — showed "intersex" features (Bhandari, 2008). Endocrine-disrupting chemicals (EDCs) in municipal effluents directly affect the sexual development and reproductive success of fishes, but indirect effects on invertebrate prey or fish predators through reduced predation or prey availability, respectively, are unknown. A recent report from the U.S. Geological Survey (USGS) found that birth-control hormones excreted by women, flushed into waterways and eventually into drinking water can also impact fish fertility up to three generations after exposure. Endocrine-disrupting chemicals with estrogenic activity (e.g., alkylphenols) have been detected in coastal Japan. Horiuchi et. al., 2005 determined estrogenic activity in extracts of river water, seawater, sediments, and sediment cores from Tokyo Bay by in vitro gene expression assay. Fifty-one of 57 extracts had some estrogenic activity. E2 equivalents (ng E2 equivalents per gram dry weight or per liter above the limit of detection) in river water samples ranged from 0.70 to 4.01 ng/ L; in seawater samples from 0.34 to 2.52 ng/L; and in surface sediments from 2.07 to 12.1 ng/g. The relationship between salinity and estrogenic activity in water samples suggested that fresh water is one source of environmental estrogens in Tokyo Bay. Over the past 12 years, male smallmouth and largemouth bass throughout the country, including the Potomac River basin in the Chesapeake Bay region, have switched sex, developing ovaries where their testes should be, and the two disruptors are prime suspects. (Ricky Carioti/ Washington Post, 2015). Male and female pearl dace (*Margariscus margarita*) captured after EE2 additions began contained up to 4,000-fold higher concentrations of the egg yolk precursor vitellogenin than fish captured from the same lake before the EE2 additions or when compared to fish from reference lakes. Edema in the ovaries, inhibited development of testicular tissue, intersex, and histopathological kidney lesions were all evident in fish exposed to EE2. Some indications that EE2 exposure affected in vitro steroidogenic capacity of the ovaries and the testes existed, although results were not always consistent between years. Pearl dace abundance was similar in the lake treated with EE2 and the reference lake. A trend exists toward a reduced overall population of pearl dace from the treated and reference lakes, as do indications that young-of-the-year size classes are less abundant in the EE2-treated lake. Biochemical and histopathological

impacts observed in fish exposed to EE2 in this study have not yet been linked to clear population level impacts in pearl dace. Monitoring of these populations is ongoing(Palace et. al., 2014). Endocrine-disrupting chemicals (EDCs) in municipal effluents directly affect the sexual development and reproductive success of fishes, but indirect effects on invertebrate prey or fish predators through reduced predation or prey availability, respectively, are unknown.

Conclusion

It can be concluded that birth control pills in municipal waste water cause negative impact on male fish population. This review suggests that the EE2 causes endocrine distruption in fishes. Most of the rivers are now polluted due to discharge of sewage. In India this work has not been done till date but a chance of pollution is not neglected. Treatment of water is not easy because it is expensive and there is low chances of purity.

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