Environmental Sciences Europe
a SpringerOpen Journal

RESEARCH Open Access

34 years of investigation in the Rhine River at Ludwigshafen, Germany – trends in Rhine fish populations

Sascha Pawlowski[1*], Juergen Jatzek[2], Thomas Brauer[2], Katja Hempel[3] and Roland Maisch[2]

Abstract

Background: The Rhine-Neckar region is one of the most urbanised areas along the Rhine River and the world's largest industrial site, BASF SE, is located here at Ludwigshafen am Rhein. When the water quality dropped to its lowest values in the 1970s, BASF SE implemented a sewage treatment plant at its production site. In addition, electrofishing at this site has been carried out at regular intervals since 1976 in order to monitor changes in the fish population, whereby clear trends in both fish diversity as well as abundance have been noted.

Results: Especially rheophilic fish species such as asp *Aspius aspius* (L.), barbel *Barbus barbus* (L.) and nase *Chondrostoma nasus* (L.) which were rarely found during the first catching period (1976 – 1980) were then caught in increasing numbers and abundance. Starting with 6 to 8 fish species per catch in the 1970s, the number has raised to 25 fish species (including 4 neozoans) in recent catches. Overall 31 species have been monitored in the last 34 years.

Conclusions: This indicates good river water quality and an increase in fish biodiversity along the BASF site at Ludwigshafen over the last three decades. However, focussing on the past decade, new invasive fish species such as the sunfish *Lepomis gibbosus* (L.) and three gobies have been found in high densities along the river banks which might have an impact on the fish population in this area in the future.

Keywords: Rhine fish population, Electrofishing, Water quality, Fish species dynamics

Background

The Rhine is one of the most "mainstreamed" rivers in Europe, balancing several aspects including transport, drinking water supply, cooling water, energy reservoir, as well as being used for effluent disposal, and providing leisure activities such as fishing (Figure 1). The former structure of the river itself had been dramatically changed as a result of the invasive canalisation project by Tulla at the beginning of the 19th century, affecting the entire river from Rheinfelden (Switzerland) to Rotterdam (The Netherlands) [1]. Based on historical fishing data, it can be concluded that this structural change also influenced the fish fauna of the Rhine [2-9]. However, especially after the Second World War, a drop in both fish species number and abundance was observed due to the contamination of the river as a consequence of increasing industrialisation and further urbanisation of the Rhine valley [10-12]. In the 1970s water quality dropped to its lowest levels resulting in the implementation of various Rhine restoration programs, including the re-introduction of endangered and/or temporarily extinct fish species [13-16]. One of the commitments was the installation of the world's largest industrial sewage treatment plant (approx. 110 million m^3 waste water per year, equivalent to a city of approx. 3 million inhabitants) located at the BASF SE site in Ludwigshafen am Rhein (Rhine-Neckar Metropolis Region, Germany) [12,17]. Along with this project, electrofishing was carried out by BASF SE at regular intervals in order to investigate potential trends in fish populations close to the industrial site in Ludwigshafen [18]. Starting in 1976, the species caught have been identified, their relative abundance determined and their overall health status observed.

* Correspondence: sascha.pawlowski@basf.com
[1]Department of Product Safety, BASF SE, GUP/PA, Z 570, Ludwigshafen 67056, Germany
Full list of author information is available at the end of the article

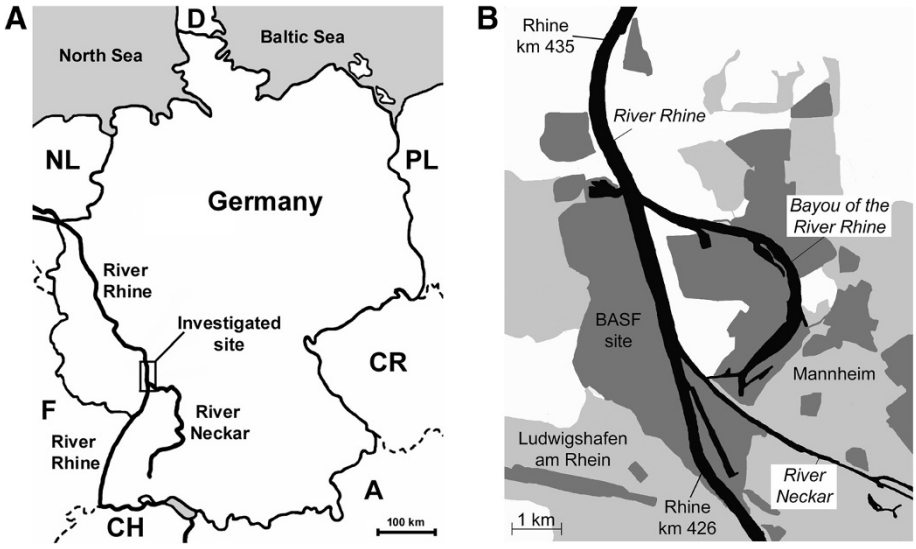

Figure 1 Schematic map of Germany (a) and the Rhine-Neckar region (b) including the Rhine River site investigated (km 426 – 435). Black – aquatic areas; dark grey – industrialised area; light grey – urbanised area; white – agricultural, nonurban and forest area.

Results

Number of fish species caught per sampling

Starting in December 1976, 6 fish species were found along the Rhine site at BASF SE Ludwigshafen (Figure 2). From 1981 until 1990 the number of fish species found varied from 8 in 1983 to 11 fish per sampling in 1985. In the next decade, a minimum of 10 fish species per sampling (1992) and a maximum of 15 fish species per sampling (1995) were found. From 2001 until 2010 the number of fish species found further increased and ranged from 12 (in 2002) to 17 (in 2006). There is still a high variation in fish species caught during the different samplings. Overall there was a statistically significant ($p \leq 0.05$) increase in the species number over the observation period as proven by linear regression by time (SAS procedure Proc Reg).

Number of fish species found per time period

During the four time periods, the total number of fish species found increased (Figure 3). During the first decade 15 fish species were located at the BASF SE site (Table 1). During the second period up to 22 species

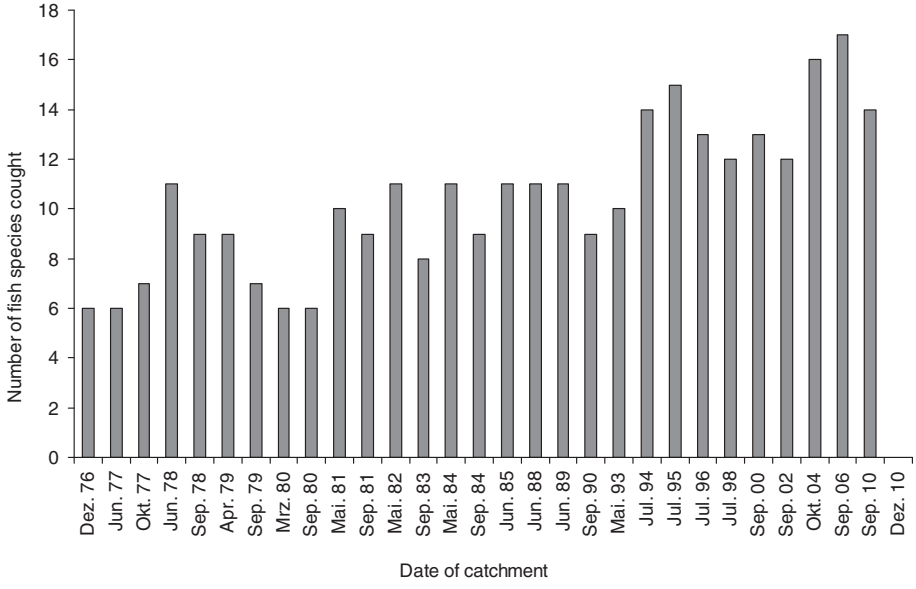

Figure 2 Number of fish species caught on each electrofishing occasion along the BASF SE site at Ludwigshafen am Rhein.

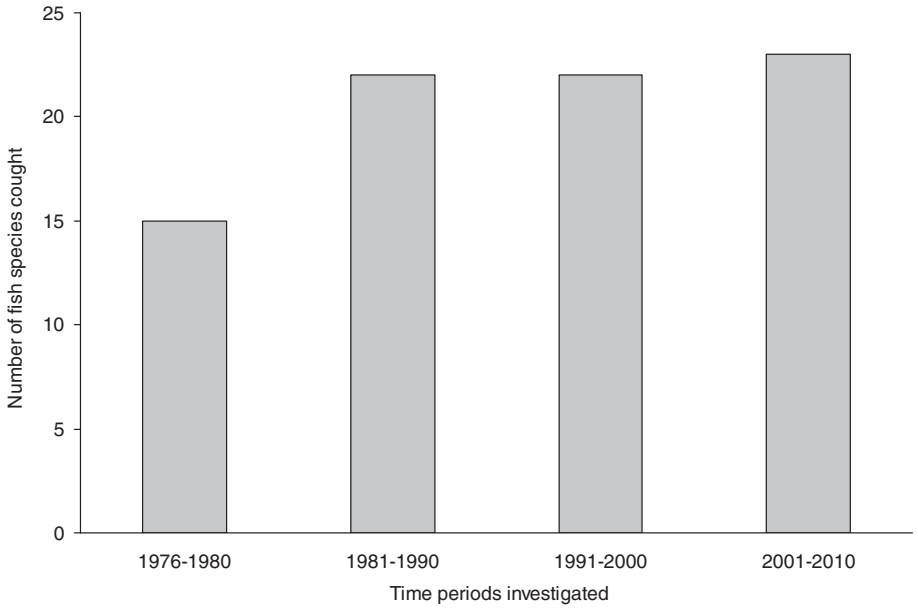

Figure 3 Total number of fish species caught during the four sampling periods from 1976 until 2010 along the BASF SE site at Ludwigshafen am Rhein.

were found. In the third period the maximum number of fish species remained the same (22). However, three further fish species were first observed in this river section and three other ones were then absent. In the most recent decade, 24 fish species including three gobidae and one cyclostoma (namely *Lampetra planeri* (Bloch 1784)) were found. Again, some species, such as *S. trutta forma fario, S. trutta trutta, L. lota* and *C. taenia,* were absent in this electrofishing period.

Relative abundance and trends in the fish community

During the 34-year investigation, 5 out of 31 fish species were found at each sampling period (100 % abundance), i.e. *C. carpio, P. fluviatilis, R. rutilus, S. erythrophthalmus* and *A. anguilla* (Table 2). Fish species such as *A. aspius, B. barbus, C. nasus, S. glanis* and *G. cernua* were absent within the first observation period, but their relative abundance constantly increased to 75 (*C. nasus*) and 100 % (all other four species), up to 2010. For *S. trutta fario, S. trutta trutta* and *E. lucius* a decrease in species abundance could be observed during the entire catching period. All other species were either commonly found (such as *A. anguilla* or *R. rutilus*) or only found occasionally (such as *L. lota* or *C. taenia*) and thus, no specific trends in relative abundance could be determined.

During the first catching period, nine endangered fish species listed in the red list of Germany and the Rhineland Palatinate were found at this site, whereas in the last decade of catches 14 fish species were found (Table 1). Only *S. trutta fario, S. trutta trutta* and *C. taenia* were absent.

Statistical evaluation as shown in Table 2 revealed a statistically significant increase for the following species: *B. barbus, S. cephalus, L. leuciscus, G. cernua, C. nasus, A. aspius, L. gibbosus,* and *S. glanis.* A statistically significant decrease was only seen for *E. lucius.* For all other species no statistically significant changes were observed.

Overall health status

Visual assessment of the overall health status, any external abnormalities, and infection status with ectoparasites showed no remarkable results [19]. Only sporadic nematodes infections of the airbladder of the eel in 1988 and 1989 are worth noting.

Discussion

The investigation of the fish population along the Rhine from kilometre 426 to 435 (including the BASF SE industrial site) over the past 34 years shows an overall increase in both species number and abundance. In fact, the fish biodiversity has increased during the whole observation period. Especially rheopilic species, such as *A. aspius, B. barbus, S. cephalus* and *C. nasus*, which were either absent or only rarely observed in the first sampling period, are now found regularly in the Rhine along this site. Although water flow has not changed essentially during this time, water quality has improved dramatically in recent years, thus improving the habitat of endangered fish species. In fact, both the nutrition load (such as nitrates and phosphates) and the concentration of organic contaminants decreased due to installation of sewage treatment plants (both municipal and

Table 1 Species list and relative abundance of fish found during each of the four sampling periods at the BASF site at Ludwigshafen am Rhein

Fish species and cyclostomes		Conservation status Germany	Conservation status Rhineland-Palatinate	Frequency of occurrence [%]			
Common name	Scientific name			1976-1980 (n=9)	1981-1990 (n=10)	1991-2000 (n=6)	2001-2010 (n=4)
Asp[1]	*Aspius aspius*	-	2	0	20	100	100
Barbel[1]	*Barbus barbus*	2	2	0	10	83	100
Bighead goby[2]	*Neogobius kessleri*	-	-	0	0	0	25
Bleak	*Alburnus alburnus*	-	-	78	100	67	75
Bream	*Abramis brama*	-	-	100	100	100	50
Brown trout	*Salmo trutta fario*	-	2	33	10	17	0
Burbot	*Lota lota*	2	2	0	20	34	0
Carp[1]	*Cyprinus carpio*	2	-	11	20	34	75
Catfish[1]	*Silurus glanis*	2	3	0	40	100	100
Chub[1]	*Squalius cephalus*	-	-	33	90	100	100
Dace	*Leuciscus leuciscus*	3	-	0	10	0	75
Eurasian ruffe[1]	*Gymnocephalus cernua*	-	-	0	40	83	100
European brook lamprey	*Lampetra planeri*	2	2	0	0	0	50
European bullhead	*Cottus gobio*	2	2	0	0	0	25
European eel	*Anguilla anguilla*	3	4	100	100	100	100
European perch	*Perca fluviatilis*	-	-	100	100	100	100
Gudgeon	*Gobio gobio*	-	3	11	60	50	50
Nase[1]	*Chondrostoma nasus*	2	2	11	10	34	75
Pike[3, 4]	*Esox lucius*	3	2	78	60	34	25
Pikeperch[2]	*Sander lucioperca*	-	4	56	60	67	75
Prussian carp, gibel carp	*Carassius gibelio*	-	-	0	20	0	0
Roach	*Rutilus rutilus*	-	-	100	100	100	100
Round gobi[2]	*Neogobius melanostomus*	-	-	0	0	0	25
Rudd	*Scardinius erythrophthalmus*	-	4	100	100	100	100
Sea trout	*Salmo trutta trutta*	2	1	33	10	17	0
Silver bream	*Blicca bjoerkna*	-	-	0	0	0	25
Spined loach	*Cobitis taenia*	2	2	0	0	17	0
Spirlin	*Alburnoides bipunctatus*	2	2	0	0	0	25
Sunfish[2, 3]	*Lepomis gibbosus*	-	-	0	0	50	25
Tench	*Tinca tinca*	-	-	33	30	0	0
Tubenose goby[2]	*Proterorhinus marmoratus*	-	-	0	0	0	25

[1] Numbers increased during the observation period.
[2] Neozoa.
[3] Found mainly at the lentic harbour side.
[4] Numbers decreased during the observation period.

industrial) along the Rhine [20,21]. Furthermore, the implementation of the large industrial sewage treatment plant at BASF SE Ludwigshafen may have had a major impact on water quality.

The reduction in organic carbon load also had a positive effect on the oxygen content of the water, thus increasing the survival rate of the more oxygen-dependent fish species historically typical of this section of the Rhine (socalled barbel region). In the early 1970s, oxygen concentrations were close to the lower limit for fish survival, resulting in a reduction of the macroinvertebrate species to about one third of the original 80 species known to be native in this area [22]. So more robust fish species like *R. rutilus/S. erythrophthalmus (*these species were not exactly differentiated at that time*), A. brama* and *A. anguilla* were the predominant species in this section of the Rhine. This is in line with previous observations from the late 1960s when *R. rutilus* and *A. brama* were found to be most abundant and fish species such as *B. barbus, S. cephalus* and *S. lucioperca* were rare in this area [23]. However, by the early 1990s, water quality measurements showed oxygen concentrations close to 100%

Table 2 Statistically significance of increases or decreases in numbers over the entire observation period

Fish species	Statistically increased over observation period	Statistically decreased over observation period
Aspius aspius	yes	no
Barbus barbus	yes	no
Chondrostoma nasus	yes	no
Esox lucius	no	yes
Gymnocephalus cernua	yes	no
Lepomis gibbosus	yes	no
Leuciscus cephalus	yes	no
Leuciscus leuciscus	yes	no
Silurus glanis	yes	no

Wald Chi Square test (logistic regression) with Significance level of 5%.

saturation with corresponding NH_4-N, NH_3-N and P-total concentrations of < 0.5, < 4 and ≤ 20 mg/L, respectively [21]. The improvement in water quality around the BASF SE site is in line with the overall improved water quality observed at other sites along the Rhine [14,24].

Rheophilic fish have increased in the past two decades but were low in numbers for the first 15 years [18]. In fact, taking in to account the whole time period of investigation this increase in the number of species found was highest between the first two decades (1976 – 1980 versus 1981 – 1990). Based on the historic data 48 fish species were found to be indigenous for the river Rhine, however recatchment analysis revealed that only 42 fish species were found in 1986/1987 [25]. Additionally, this number of fish species refers to the whole river section including also estuaries and bayous and is therefore not restricted to the main river section investigated [26]. This would explain why fish species such as the *Coregonus* spp. L. (2 species), *Osmerus eperlanus* L., *Misgurnus fossilis* L., *Rhodeus amarus* (Bloch), *Gasterosteus aculeatus* L. and *Pungitius pungitius* L. were listed as indigenous for the river but should not be considered to inhabit this specific area [27].

Those fish species found are representative of the Rhine, and can be found further upstream at the socalled Hochrhein (from Lake Constance to Basel, Switzerland), thus confirming the good water quality and high oxygen content [28]. Nevertheless it cannot be excluded that rheophilic fish species has recovered due to other reasons (e.g. fish passes). The wide variety of fish species caught at each sampling, however, can be explained by the method used as it is limited to the surface and river bank areas and thus strongly dependent on the given water flow conditions, which beyond water temperatures (high or low water; see Table 3) [23,29,30]. Despite these limitations, a clear trend in fish population dynamic can be seen during the past decades of investigation.

Table 3 Corresponding water levels and water temperatures on the month of electrofishing

Date of fishing	Water gauge [m] Mannheim	Water temperature [°C] Ludwigshafen
Dec 1976	n. d.	5.5
Jun 1977	3.82	17.1
Oct 1977	2.17	14.1
Jun 1978	4.35	16.8
Sep 1978	2.91	17.5
Apr 1979	3.50	9.7
Sep 1979	n. d.	18.5
Mar 1980	n. d.	7.9
Sep 1980	n. d.	17.8
May 1981	n. d.	13.9
Sep 1981	n. d.	18.2
May 1982	3.98	14.0
Sep 1983	2.92	18.5
May 1984	2.19	12.9
Sep 1984	3.10	17.5
Jun 1985	4.20	21.5
Jun 1988	4.12	17.9
Jun 1989	2.13	19.9
Sep 1990	1.67	19.0
May 1993	2.91	17.7
Jul 1994	2.90	23.9
Jul 1995	3.98	22.5
Jul 1997	2.39	19.7
Jul 1998	2.63	21.7
Sep 2000	2.80	19.0
Sep 2002	n. d.	18.6
Oct 2004	n. d.	15.8
Sep 2006	n. d.	20.1
Sep 2010	3.41	18.3

n. d. – no data available.

In addition to the increase in species numbers, several other observations on fish fauna have been made which are not necessarily linked to the water quality improvements in this area in particular. Restocking measures which were carried out during the past tenth of years for *S. salar, S. trutta* and *A. anguilla* might also have and direct (species affected) or an indirect impact (predator) on the river rhine fish populations [15,31,32]. Nevertheless, the number of both *S. trutta fario* and *S. trutta trutta* has decreased over the past 34 years and are absent nowadays. These two species, however, have always been rare as they are more likely to be found in the lotic tributaries of the river (socalled trout region) than in the Rhine itself.

Furthermore, the *S. glanis, G. cernua* and *C. carpio* are not typical of the barbel region as they are more native

to lentic habitats such as lakes and the lowlands of the Rhine. The increase in their relative abundance might be explained either by the direct connection to lentic harbours and bayous or especially by the rocky structure of the river bank which provides an excellent hiding habitat for at least the *S. glanis* and *G. cernua*. In addition, since the number of catfish was low in the Rhine in the past, this species was restocked several years ago and is now to be found in great numbers. Considering other species found more or less frequently over the 34-odd years of investigation, it should be mentioned that fish usually migrate along the river but show a preference for a specific river section (as indicated by trout or barbel or bream region). Thus, it is probable that a fish species can be found in a river section where they are not usually abundant. The *E. lucius,* for example, was usually caught in the lentic harbour at the BASF SE site which in fact is the more appropriate habitat than the river itself und thus their relative low abundance compared to *P. fluviatilis* and *S. lucioperca* is not unexpected [27].

Although the number of fish species has risen over the past 34 years, the species list differs from the one drawn up 100 years ago, since some anadromous fish such as the allis shad *Alosa alosa* (L.) and common sturgeon *Acipenser sturio* L. have been extinct for more than 60 years due to former river contamination (at the beginning of industrialisation), barricades along the length of the river and intensive fishing [33]. For the Atlantic salmon *Salmo salar* L. several reimplementation programmes were established in the late 1990s to restock populations [34]. This species has still to be found during the electrofishing procedure as the method used is limited to catches along the river banks rather than the middle of the river, and the number of returning *S. salar* is still low [30].

However, some new invaders have been observed: the North American sunfish *L. gibbosus* has been found in the area since the early 1990s, originating from either sport fisheries (1st invasive period) or pet shop trading (2nd invasive period). In 2010, high population densities of another invasive group of fish, the pontocaspian gobies (Fam. Gobidae), were noted. Although the exact species has not been determined, it is very likely that they found their way into the Rhine via the Rhine-Main-Danube Canal since at least the *Proterorhinus marmoratus* (Pallas 1814) was found in the River Main in the late 1990s [35]. Like the invasion of Danubian/Caspian fish species also the Rhine macroinvertebrate fauna is highly affected by Danubian species [36].

Both sunfish and gobies are strong predators especially of young fish, but at least for the latter species there is little information on their possible impact on the Rhine fish community to date.

For some fish species found in the past one or two catching periods, such as the *A. bipunctatus, L. leuciscus* and the cyclostoma *L. planeri,* no definitive population trend analysis is currently available since the time period of their abundance is too short. However, as they are "native" to the barbel region it is very likely that their numbers will rise in future.

Overall, even in the highly industrialised river section at Ludwigshafen am Rhein, the Rhine fish population along with the water quality do not markedly differ from other comparable river sections, showing a good recovery over the past 34 years of investigations.

Conclusions

Overall, the water quality of the Rhine has improved since the 1970s as indicated by the number and species of fish collected by electrofishing in the period of investigation. The improvement of water quality is also enhancing the biodiversity of fishes in this river. Since the water quality has almost reached the optimum which can be expected for this river section (water quality index of about 2.0 – 2.3) the number of fish species found may likewise have reached maximum levels for this water quality. However, current trends indicate a shift towards invasive Neozoa so there might be a further biological impact on the Rhine fish population in future. Therefore, regular electrofishing is considered to be a valuable tool not only to monitor water quality but also to constantly investigate the fish populations along the BASF SE site with respect to new invaders and potentially new species.

Methods

Samples were taken by electrofishing at regular intervals between 1976 and 2010 along the left bank of the Rhine from Gate 6 of BASF SE (Rhine km 426) to Rheindürkheim (Rhine km 435, Figure 1).

Fish were caught using a small boat with equipment on board [18]. Briefly, a 500 volt (12 ampere) direct current generator was connected to an anode dip net (diameter of about 40 cm; mesh width about 1 cm) as used to catch the fish. Fishing takes places about 3 to 5 m from the western shore of the river Rhine at about 1 m water depth.

Species were determined and their local abundance was assessed semiquantitatively. Since the species found vary for each sampling, the total number found was clustered into four time periods (until 1980 [period 1], 1981 – 1990 [period 2], 1991 – 2000 [period 3], and 2001 – 2010 [period 4]) in order to investigate trends in abundance over the 34-year sampling period. All fish were also checked visually for overall health status, external abnormalities, and infection with ectoparasites.

From 1976 to 2010, 29 samplings were taken. In some years between 1976 and 1984 the fish were caught in the spring and autumn in order to determine any differences in the number of species between the two seasons. From 1988 on, electrofishing was mostly executed in autumn. Monitoring was introduced by BASF SE as a commitment to responsible care and also to measure the effects of enhanced water cleaning measures and improved treatment procedures at the site itself [37].

Statistical methods

For the binary response (species present or not present) increase or decrease in the occurrence by time was tested using SAS procedure Proc Logistic.

This analysis was done separately for each species using logistic regression [38]. The Wald Chi Square test was used to test for an increase or decrease by time. For the number of species a linear regression by time was done using the SAS procedure Proc Reg [39]. It was tested if there is an increase or decrease by time.

Competing interests

The authors declare that they have no competing interests.

Authors' contributions

SP was part of the project team carried out the last electro fishing in 2010, data collection, literature review and drafted the main sections of the manuscript. JJ was responsible for the organisation of the electro fishing carried out during the time of investigation starting in 1987 and was also part of the experimental project team. Furthermore, he was involved in the literature review and contributed to the historical data of the past electrofishing occasions. TB was part of the experimental project team during the last electrofishing occasion in 2010. KH was part of the experimental project team during the last electrofishing occasion in 2010 and responsible for coordination of statistical analysis of the results and the critical review of the manuscript. RM was reviewing the last version of the manuscript and gave additional input in that version prior to its internal release. All authors read and approved the final manuscript.

Authors' information

All authors, but Katja Hempel are working at the Department of Products Safety at BASF SE Ludwigshafen as experimental and regulatory ecotoxicologists and toxicologists, respectively. Katja Hempel is now working as a Toxicologist for Abbott GmbH & CO. KG in the Department of Global Preclinical Safety at Ludwigshafen.

Acknowledgements

The authors gratefully thank G. Kuhn, Karlsruhe, K. Kuhn, Leimersheim, T. Oswald, SGD Neustadt, U. Pagga, Ludwigshafen and R. Bias, Wachenheim and their coworkers for their collaboration and support during the various electrofishing activities and Martina Dammann for statistical analysis of the data.

Author details

[1]Department of Product Safety, BASF SE, GUP/PA, Z 570, Ludwigshafen 67056, Germany. [2]Department of Product Safety, BASF SE, Ludwigshafen 67056, Germany. [3]Abbott GmbH & CO. KG, Global Preclinical Safety, GPRD, Ludwigshafen 67061, Germany.

Received: 25 April 2012 Accepted: 8 October 2012
Published: 17 October 2012

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doi:10.1186/2190-4715-24-28
Cite this article as: Pawlowski *et al.*: 34 years of investigation in the Rhine River at Ludwigshafen, Germany – trends in Rhine fish populations. *Environmental Sciences Europe* 2012 **24**:28.