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Nitrogen isotopes in herbaria document historical nitrogen sewage pollution in the Mersey Estuary, England†

Freya C. Alldred, *[a] Darren R. Gröcke, *[a] Samuel E. Jackson [b] and Geraldine Reid [c]

A macroalgae (seaweed) herbarium nitrogen isotope ($\delta^{15}N$) record is produced for the River Mersey and Liverpool South Docks (England) between 1821 and 2018. A modern macroalgae $\delta^{15}N$ record was also produced from September 2022. The herbaria $\delta^{15}N$ record shows a stark difference from 1821 to the present. Lower $\delta^{15}N$ in the early 1800s is attributed to agricultural and raw sewage pollution. From 1970 to the present the herbaria samples record very elevated $\delta^{15}N$ values – peaking in 1978 at +31‰. The 1989 Water Act and privatisation of water companies in the UK had limited impact on the herbarium $\delta^{15}N$ record but indicated a dominance of sewage nitrogen in the River Mersey. Macroalgae $\delta^{15}N$ has become even more elevated since the last herbaria sample in 2013. The herbaria and modern data record some of the highest seaweed $\delta^{15}N$ values (and therefore, sewage nitrogen pollution) recorded to date. This study highlights a novel use of herbaria macroalgae to document past changes in nitrogen pollution in estuarine environments. More poignantly it highlights that the River Mersey – Mersey Estuary is heavily polluted with sewage nitrogen and requires immediate action to resolve this environmental issue.

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Environmental significance

Stable nitrogen isotope ratios ($\delta^{15}N$) in macroalgae are an under-utilised tool in the UK for studying coastal and estuarine pollution. Nitrogen loading from effluent and industrial sources produce distinct nitrogen isotope signatures. Herbaria are an untapped resource for understanding anthropogenic modification in the environment. This study uses macroalgae herbaria to identify broad nitrogen pollution changes over the past 200 years for the Mersey Estuary, UK. This is the first study of its kind to use herbarium macroalgae to investigate nitrogen pollution. Herbaria and modern macroalgae $\delta^{15}N$ indicate that the Mersey Estuary has been dominantly affected by sewage for over 200 years.

Introduction

Wastewater discharge in UK rivers and coastal environments is becoming more frequent which is leading to a decline in water quality.[1–3] In February 2024, there are *no* rivers in England that have good overall health status and only 15% of rivers are of good ecological health status.[1–3] This is in stark contrast to Scotland which has ~57% of its rivers with good (or better) overall health status.[4] Policy changes to permit combined sewer overflows (CSOs) to discharge raw sewage at times of peak flow have been detrimental to the ecological health of UK rivers.[5] Many wastewater treatment facilities are inadequate to cope with current population levels and have had minimal investment for decades, causing the number and frequency of CSOs to increase.[5] This environmental crisis was graphically highlighted in the BBC Two documentary, *Our Troubled Rivers*.[6] Discharge during periods of low flow in rivers increases the residence time of the pollutants, enhancing the detrimental impact it has on the environment. Excessive nutrient loads are responsible for phytoplankton, algal and macrophyte blooms, which subsequently reduce oxygen contents leading to eutrophication of water bodies.[7–9] The current environmental issues in England rivers are exacerbated by budget cuts in the Environment Agency, which has hindered their ability to monitor, designate and prosecute water companies over illegal discharges of sewage.[5,10,11] It is also hampered by an unwillingness from privatised water companies to openly share data on discharge amounts and dates, resulting in the public taking on the responsibility to report and monitor wastewater discharges.[12,13] There is widespread concern nationally over the effects that wastewater (*i.e.*, raw and treated sewage) release has had on riverine and coastal environments.

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Sewage effluent that reaches the coastal ecosystem can be identified through nitrogen isotope ($\delta^{15}N$) analysis of organisms living in that environment (*e.g.*, macroalgae, mussels and fish).[14–17] Macroalgae (seaweed) is less-frequently used as a bio-monitor to trace nitrogen sources, especially in the UK.[17,18] Macroalgae takes up nitrogen (*e.g.*, ammonium and nitrate) with minimal nitrogen isotope fractionation and therefore, can be used to discriminate the nitrogen source in the marine environment.[19] Different nitrogen sources (*e.g.*, fertilisers, raw and treated sewage) have distinct isotopic averages. Treated sewage effluent is often identified through $\delta^{15}N$ values greater than +7‰ in macroalgae.[17,20,21] On the other hand, nitrogen pollution derived from industrial chemical processes (*e.g.*, artificial fertilisers) produces $\delta^{15}N$ values near the atmospheric nitrogen value, ≈0‰.[22–24] Although there are numerous studies that have used macroalgae $\delta^{15}N$ to identify sewage pollution, there are only a handful of studies that exist for the UK despite the ongoing sewage pollution crisis.[17,21,25,26]

Museum herbarium collections contain a vast amount of ecological and environmental information that is often underutilised in the study of recent anthropogenic change.[27,28] Only recently have researchers considered herbaria as a tool to track biogeographical, environmental, and climatological changes.[28–30] Although herbaria relate to all forms of dried materials, such as vascular plants, macroalgae, bryophytes, lichens, and fungi, 82% of current research has focussed on accessing vascular plant collections.[29] Herbaria studies have dominantly been used to record population densities, distribution, and organism morphology to infer environmental conditions, but more recently this has extended to include DNA sequencing and chemical analyses.[27,29,31] Macroalgae herbaria are starting to be used to investigate the marine environment although they remain underutilised on samples prior to the 20th century.[29,32,33] Recently, Miller *et al.*[32] used macroalgae herbaria and nitrogen isotopes to reconstruct past upwelling trends along the Californian coast – this approach was adopted since that is the dominant environmental mechanism affecting nitrogen isotopes in that region.

In this study, we used macroalgae specimens collected and stored in the herbarium at the World Museum, National Museums Liverpool to reconstruct historical nitrogen pollution in the Mersey Estuary and Liverpool Docks. Herbaria specimens from the Mersey Estuary were assessed for their suitability for destructive sampling. Only those where there was adequate material available to have a portion removed without damaging the scientific integrity of the specimen for future research were used in this study. Very delicate small specimens, for example, were avoided and for pre-1900 specimens only one sample per year was permitted for sampling: again, to preserve the herbarium collection for future research. The collection has been generated relatively consistently from the same region between 1821 and 1860 and from the 1960s to the present day. Sampling gaps occurred during World War I and II, and may be a common artefact in herbarium records around Europe for this time interval. Although this is unfortunate from a scientific perspective, the herbaria record available will still allow us to reconstruct changes in nitrogen pollution from the Industrial Revolution to the modern sewage era. The River Mersey and Mersey Estuary has witnessed significant anthropogenic changes over the past 200 years and thus, is an ideal natural setting to assess the use of macroalgae $\delta^{15}N$ from herbaria as a proxy for reconstructing historical nitrogen pollution.

The River Mersey and estuary

The River Mersey has a catchment area of ~1800 mi^2 and includes Manchester, Lancashire and Cheshire in the north-west of England.[34] It flows for 69 miles before widening into the Mersey Estuary that stretches for almost 16 miles (Fig. 1).[35] The large metropolitan city of Liverpool is located at the mouth of the River Mersey (Fig. 1), where large tidal ranges (>10 m) cause strong currents and large sand banks.[36] The Mersey Basin has grown in population size from ~500 000 (1821) to >5 million (2021); this estimate includes other nearby population centres such as Manchester, Liverpool, and Salford.[37–39] This region is serviced by the private water company, United Utilities Group PLC. The River Mersey and Mersey Estuary have had a long history of pollution and poor water quality since the early 1800s.[36,38] Nitrate plumes originating in the River Mersey have seriously impacted Liverpool Bay since the 1960s.[34,40] Public outcry in the 1980s incentivised the launch of the Mersey Basin Campaign.[38,41] The aim was to clean up the polluted River Mersey after it was described as an "affront to the standards a civilised society should demand" by the then Secretary of State for the Environment, Lord Heseltine.[42] Unfortunately, pollution issues still exist in the River Mersey. For example, data extracted from The Rivers Trust Sewage Map[43] show that in 2021 the River Mersey catchment area experienced over 212 000 h of effluent discharge prior to processing through 12–24 h (for reference, a calendar year = 8760 h).

The River Mersey and Mersey Estuary macroalgae herbaria $\delta^{15}N$ record spans 197 years consisting of 70 macroalgae specimens collected between 1821 (*Enteromorpha compressa*, Liverpool) and 2018 (*Polysiphonia stricta*, Queens Dock)—including the time gap previously mentioned (see the ESI†). Many different macroalgae species have been used to generate the $\delta^{15}N$ record, because a range of specimen types are collected for herbaria; these are often selected based on casual observation, ecological monitoring, identification and taxonomy, or more frequently because of their fragility, rarity, and beauty on herbarium paper. A brown seaweed is less often collected, such as *Fucus vesiculosus* (bladder wrack), because it is not as eye-catching as a delicate red seaweed and is very abundant around the UK coastline. The same macroalgae species are often not routinely collected for herbaria and hence, generating a long-term species-specific $\delta^{15}N$ record will not be possible. We suspect that this will be an inherent issue with many herbarium collections around the world. Irrespective of the use of different macroalgae species the $\delta^{15}N$ record produced in this study reveals consistent and significant changes over the past 200 years that can be related to societal changes and a major neoliberalism event in 1989 in the UK:[44] the privatisation of water companies.

F. vesiculosus collected from Eastham in 1978 recorded the most elevated $\delta^{15}N$ value of +30.6‰, whereas the lowest $\delta^{15}N$ value was −4.1‰ collected from Otterspool in 1968 (also *F. vesiculosus*) (see the ESI†). $\delta^{15}N$ values of macroalgae above +20‰ are very rare in the literature[45] and suggest extreme environmental conditions; in this case, we interpret these elevated $\delta^{15}N$ values as a result of

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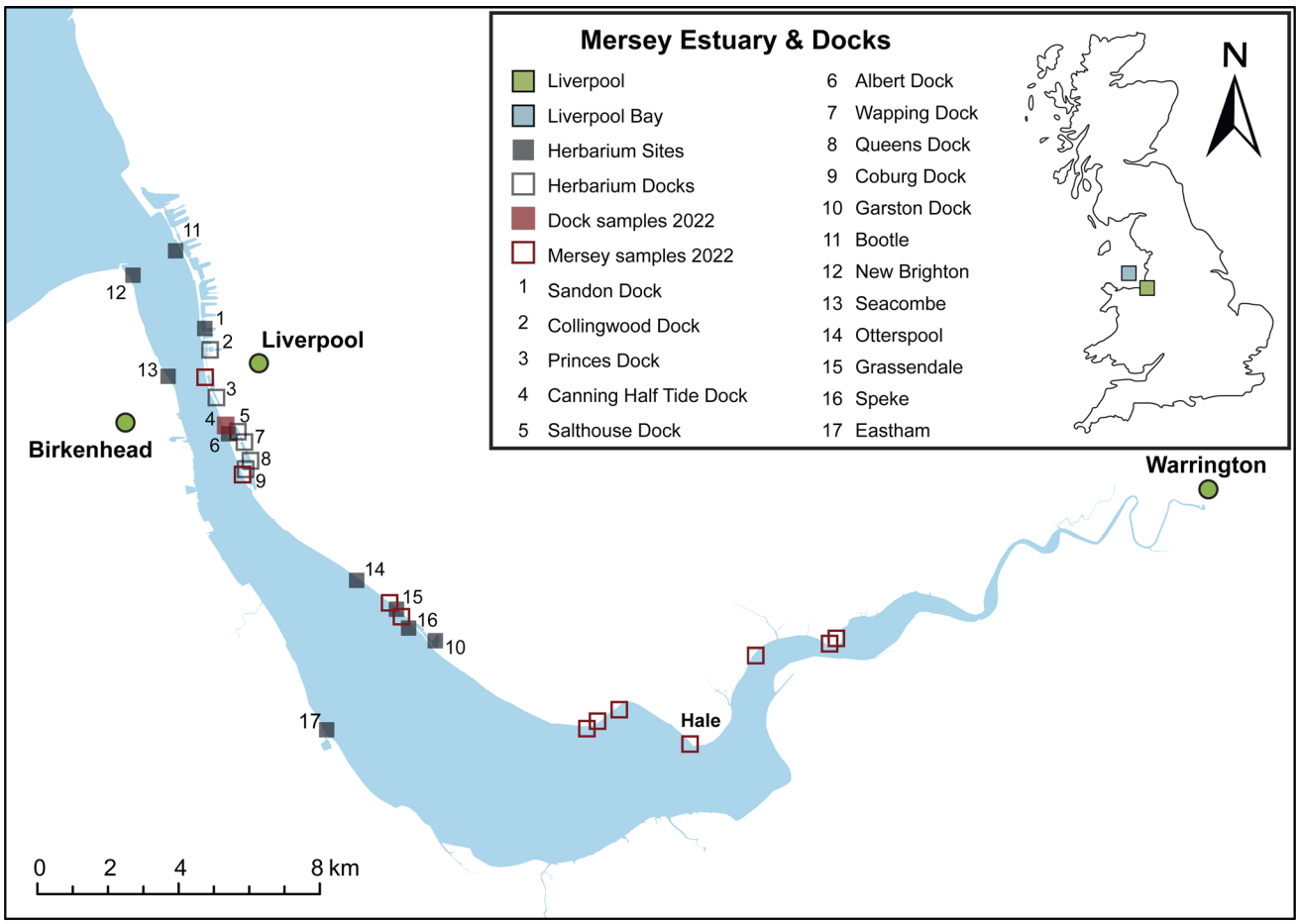

Fig. 1 Study area of the Mersey Estuary with modern sample sites (red) and herbarium sites (grey). Hale Lighthouse is located south of the village of Hale. ArcGIS Pro 3.0 was used to produce this figure.

continued, voluminous release of sewage into the River Mersey. Four herbaria samples from Birkdale (along the coastline north of Liverpool) were also analysed and exhibit a range between +2.9‰ and +9.1‰ spanning the time interval, 1936–1938; these data are not discussed any further in this study. To place the herbaria $\delta^{15}N$ record in the context of the present-day environment, a series of macroalgae samples from the Mersey Estuary and South Docks were collected in September 2022 (see the ESI†). Many different macroalgae species were collected from the South Docks and produced an average $\delta^{15}N$ value of +10.6‰ ± 3.2‰ ($n = 27$) (Fig. 2). However, macroalgae $\delta^{15}N$ from the Mersey Estuary average +16.5‰ ± 2.0‰ ($n = 50$) and +15.5‰ ± 2.7‰ ($n = 22$) for *Fucus* sp. and *Ulva* sp., respectively (Fig. 3). The herbaria $\delta^{15}N$ data from the Mersey Estuary and the South Docks will be discussed separately. Due to the limited sample size, we have grouped the herbaria $\delta^{15}N$ data into age ranges as discussed below.

The South Docks herbaria record

The South Docks in Liverpool are an interconnected system,[46] and therefore the herbaria macroalgae $\delta^{15}N$ record is being treated as a single composite record. Fig. 2 shows the herbaria South Docks $\delta^{15}N$ record between 1981 and 2018. An 1846 herbaria sample collected from Princes Dock is the only dock specimen collected during the 1800s and so this sample has been omitted from further discussion (*F. vesiculosus*, $\delta^{15}N$ =

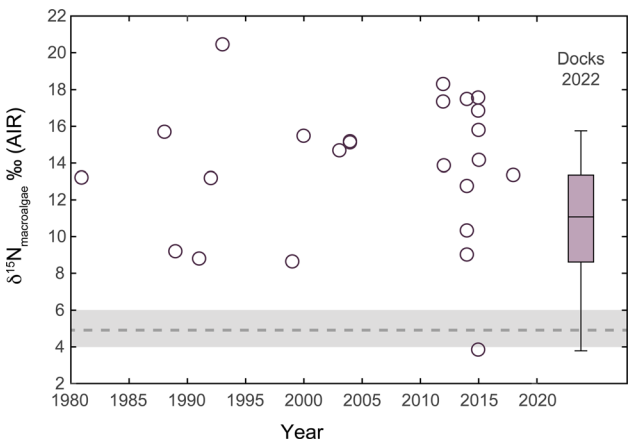

Fig. 2 Herbaria and modern nitrogen isotope record for the South Docks of Liverpool. Herbarium data points are shown in lilac ($n = 23$); the modern nitrogen isotope range is provided as a boxplot collected in September 2022 ($n = 27$). The grey box represents the "natural" isotopic range (+4‰ to +6‰) for the North Atlantic[25] and the dashed line represents the global nitrate $\delta^{15}N$ value.[51]

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+8.2‰). $\delta^{15}N$ values show no clear trend in the South Docks over the 40-year period (Fig. 2). The South Docks herbaria record has an average $\delta^{15}N$ value of +13.7‰ ± 3.8‰ (n = 23), which is significantly more elevated (p value > 0.003) than the 2022 dock average (+10.6‰ ± 3.1‰, n = 27).

Allen *et al.*[46] reported low public opinion of dock water quality in the 1990s. Efforts to limit mixing between the river and docks were obtained through the installation of a pump in 1992 to replenish water levels, thus reducing turnover rates of the dock seawater to between 6 and 12 months.[34,46,47] A reduced replenishment rate may be the reason behind dock herbaria not reflecting similar macroalgae $\delta^{15}N$ values recorded in the Mersey Estuary. The lower $\delta^{15}N$ average of the dock herbaria and modern macroalgae compared to the Mersey Estuary suggests two potential mechanisms: (a) increased industry-sourced nitrogen pollution entering the docks (*e.g.*, drains and road runoff); and/or (b) water filtering (*i.e.*, cleansing) by the presence of the bivalve, *Mytilus edulis* (blue mussel).[47] Since the mid-1980s there have been several reports of blue mussel colonies thriving in the docks; they were introduced to Graving Dock as part of a bio-filtration experiment and naturally settled in Albert Dock and subsequently throughout the South Dock system.[34,47] The duration it would take for blue mussels to filter the volume of water in Albert Dock (170 000 m^3) is calculated to be every 4 days. Scaled up to include the entire South Dock complex (1.28 million m^3) the duration to filter all that water would be ~30 days assuming that blue mussel density is consistent throughout the docks.[47,48] Nitrogen is consumed by microalgae and phytoplankton in the docks which are consumed by filtering blue mussels resulting in lower macroalgae $\delta^{15}N$ values. This is consistent with a blue mussel experiment showing that water nitrate and blue mussel tissue are more depleted in $\delta^{15}N$ when nitrate concentrations in the water are high.[49] Dock water samples analysed in this study indicate low to moderately high nitrate concentration ranges from 0.01 to 11.6 mg l^{-1} (n = 13, January–July 2023). Therefore, blue mussels, or other filtering organisms, should be considered as a natural bio-remediator in ports/harbours/docks, but they should not be considered as a solution without rectifying the cause.

The modern and herbaria macroalgae $\delta^{15}N$ record (Fig. 2) indicates that dock water quality has remained relatively consistent since its redevelopment in the 1970s and introduction of the blue mussel ecosystem in the 1980s. Whilst the dock herbaria and modern macroalgae $\delta^{15}N$ records show less elevated values compared to the Mersey Estuary, the docks still record elevated signatures (+13.7‰ and +10.6‰, respectively). Such elevated macroalgae $\delta^{15}N$ would suggest that the dock water still contains anthropogenic nitrogen sourced from raw and/or treated sewage. Since the Mersey Estuary is the primary source of seawater for the docks, a sewage nitrogen $\delta^{15}N$ signature is unavoidable.

Mersey Estuary, 1821–1863: Victorians and raw sewage

Between 1821 and 1863, herbaria $\delta^{15}N$ values ranged between +4.7‰ (*Ectocarpus granulosus* 1863 and *Enteromorpha compressa* 1853) and +21.5‰ (*Enteromorpha intestinalis* 1849) (Fig. 3). The majority of the herbaria specimens from this period have $\delta^{15}N$ values that fall between +4.7‰ and +8.8‰ (n = 14) with four other results greater than +11‰. A single specimen (*E. intestinalis*) from Bootle in 1849 recorded a $\delta^{15}N$ value of +21.5‰. This time interval records an average $\delta^{15}N$ value of +8.3‰ ± 4.1‰ (n = 18) or with exclusion of the 1849 Bootle sample, +7.5‰ ± 2.6‰ (n = 17). Fig. 3 also shows a global average deep-water $\delta^{15}N$ value of +4.8‰ and the expected range (+4‰ to +6‰) for macroalgae in the North Atlantic.[16,50,51]

Although the 1821–1863 Mersey Estuary herbaria record has a limited amount of $\delta^{15}N$ data (n = 18) and information regarding their precise sampling location and precise date, the $\delta^{15}N$ range is significantly different from the 1990–2013 (p value < 0.02, n = 13) and 2022 (p-value = 1.6 × 10^{-7}, n = 72) datasets; it is not significantly different from the 1949–1983 dataset (p value > 0.1, n = 11). During the 1800s the Mersey Basin and Mersey Estuary were dominantly influenced by industrial activities: cotton, alkali and hydrogen-chloride production, shipbuilding and transport of coal.[36,38] However, industry of this kind preceded the discovery and explosion of the use of nitrogen gas in industrial processes. For example, agricultural fertilisers made from nitrogen gas ($\delta^{15}N \approx 0‰$) were not generated until the discovery of the Haber–Bosch process and large-scale processing in 1913.[52,53] Therefore, industrial processes would not be the cause behind lower $\delta^{15}N$ values compared to the other time intervals in this study.

River pollution in England was rife during the Victorian Era.[54] The main route of sewage/wastewater disposal in the 19th century was to cast it into rivers and/or cesspits, with the latter

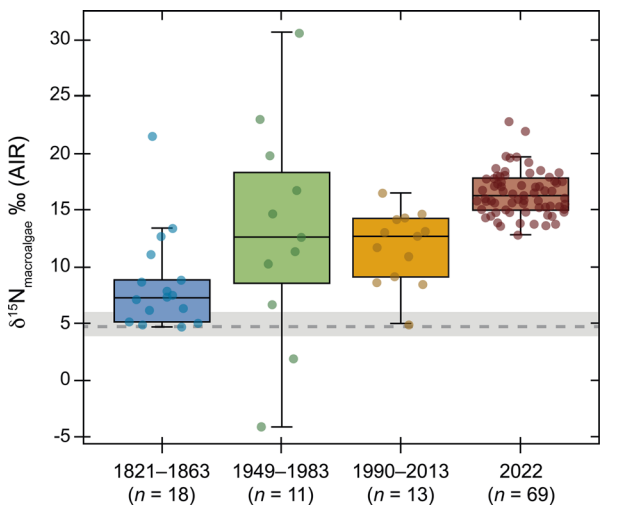

Fig. 3 Herbaria and modern nitrogen isotope record for the Mersey Estuary between 1821 and 2013 (n = 42) and 2022 (n = 69). There is a significant statistical difference between 1821–1863 and 1990–2013 and 2022. There is no statistical difference for the dataset 1949–1983 due to the large range in $\delta^{15}N$, which spans the entire range of the whole dataset of this study. The grey box represents the "natural" isotopic range (+4‰ to +6‰) for the North Atlantic[25] and the dashed line represents the global nitrate $\delta^{15}N$ value.[51]

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infiltrating into the hydrological system.[55] Between 1847 and 1858 an 80-mile-long sewer network with 48 outflows discharging into the River Mersey was constructed, focusing sewage waste in the river.[38,56] Sewage waste in England reached such a level that it caused several national cholera outbreaks;[57] the largest of these outbreaks occurred in 1849. It is interesting to observe that the 1849 cholera outbreak coincides with the most elevated $\delta^{15}N$ value (Bootle, +21.5‰) in our herbaria record and Liverpool having the highest cholera mortality rate in large towns reported for England (Liverpool, 11.3 deaths per 1000 people).[57] Even after the cholera outbreaks there are reports of major sewage issues; for example, the Great Stink of London in 1858 when the River Thames was so heavily contaminated it sparked the conception of the modern-day sewerage system.[58]

Principally, we postulate that the input of raw sewage (*e.g.*, not treated, and hence not denitrified[59]) was the cause behind the 1821–1863 $\delta^{15}N$ values recorded for the Mersey Estuary. Raw human sewage would have a similar $\delta^{15}N$ value to the diet they were consuming[60] and based on a modern-day equivalent dietary value this would represent a range between +4‰ and +8‰.[61] Although raw sewage can explain the $\delta^{15}N$ record, agricultural practices cannot be excluded.[38,56] In addition, leaching and weathering of soil-derived nitrogen from fields using organic fertilisers (*i.e.*, manure) may also be contributing to the $\delta^{15}N$ signature.[24] To our knowledge no soil-nitrogen isotope studies exist for Merseyside. In addition, no water quality data (*i.e.*, nitrate concentration) exist this far back in time for this region, unlike the River Thames, London.[39,62,63] Although the contribution of nitrogen sources is uncertain the macroalgae $\delta^{15}N$ record from 1821–1863 is most likely caused by raw sewage (*e.g.*, human and animal husbandry), and it is evident that it was different from the modern Mersey Estuary record (1990–present).

The ~100 year gap from 1863 is unfortunate since it would have been interesting to understand whether the macroalgae $\delta^{15}N$ record would follow an increasing trend or shift suddenly. There is however some information pertaining to water quality from historical records during this time interval. No "waterweeds" (*i.e.*, macroalgae) were present in the Mersey Estuary between the 1870s and 1900s which can be attributed to water pollution.[39] Furthermore, from the 1900s unregulated sewage and industrial discharges persisted until the 1950s when wastewater discharge permits were first introduced.[64,65]

Mersey Estuary 1949–1989: divergent nitrogen pollution sources

The herbaria $\delta^{15}N$ record for the 1970s is more elevated in comparison to the $\delta^{15}N$ record for the 1800s. The Mersey macroalgae herbaria record from 1949 to 1983 shows the greatest range in $\delta^{15}N$ (34.7‰) with an average value of +13.0‰ ± 9.2‰ ($n = 11$). The lowest $\delta^{15}N$ value of −4.1‰ recorded in *F. vesiculosus* occurred in 1968 at Otterspool, and the most elevated $\delta^{15}N$ value at +30.6‰ (also by *F. vesiculosus*) occurred a decade later in Eastham. It is difficult to accurately determine or constrain the cause behind the large variation in $\delta^{15}N$ between 1949 and 1983. It records one of the most elevated macroalgae $\delta^{15}N$ values (*i.e.*, caused by sewage/denitrification) ever recorded to date, but is immediately followed by a negative $\delta^{15}N$ value indicative of industrial pollution. Due to the large range in $\delta^{15}N$ for this time interval it is not significantly different from any of the age range groups assigned to this data set (see Fig. 3). Since the majority of the $\delta^{15}N$ values in this time interval are elevated, removing the lowest two $\delta^{15}N$ values causes the average value to increase to +16.2‰ ± 6.9‰. Standard processes in wastewater treatment plants cause the sewage effluent to become enriched in ^{15}N due to denitrification.[66] This time interval has very elevated $\delta^{15}N$ values in comparison to background "natural" macroalgae,[16] thus implying it is heavily influenced by nitrogen pollution created by wastewater treatment plants.[66]

Decades of historical releases of sewage and industrial pollution led to a serious decline in the health status of UK water bodies. Massive cuts in funding and spending during the 1970s and 1980s exacerbated specifically the impact of sewage pollution on the environment making it a national problem that needed addressing.[41] The Control of Pollution Act 1974[67] introduced the requirement that local stakeholders had to apply for permits to discharge sewage effluent and industrial waste or face prosecution. This was heralded as a major legal improvement although it was slow to be implemented nationally.[68] In the 1970s the Mersey Estuary was considered the most polluted estuary in the UK receiving significant wastewater inputs that subsequently led to high nitrate plumes in Liverpool Bay.[34,38,41,56,64] High nutrient fluxes as a result of sewage wastewater discharges caused biochemical oxygen demand (BOD) in the late 1960s to average 20 mg l^{-1} which is indicative of very polluted water. The enriched $\delta^{15}N$ values observed in herbaria collected for this interval corroborate the input of denitrified sewage wastewater (Fig. 3).[34] Growing public demand for improved water quality at the peak of the Mersey pollution crisis in the mid to late 1980s saw the establishment of the Mersey Basin Campaign.[41,69] At the same time public concern over environmental quality was growing across the nation.[41] The Conservative Government and the then Prime Minister, Margaret Thatcher, actioned the privatisation of water companies in England and Wales through the Water Act 1989.[70] Even then, as it is now, the privatisation of water companies was primarily focused on macro-economic policies – to inject much-needed cash into an infrastructure that had received little investment for decades.[41,71] With a failing regulatory system between DEFRA (Department for Environment, Food and Rural Affairs), the Environment Agency and Ofwat (Water Services Regulation Authority) there has been a continuous lack of investment from the water industry in keeping up with population growth and protecting the environment.[71]

Mersey Estuary, 1990–2013: the start of the sewage era

The herbaria $\delta^{15}N$ record depicted in Fig. 3 indicates that the privatisation of water companies had an impact on the macroalgae nitrogen isotope signature in the Mersey Estuary. It resulted in more stability in herbaria $\delta^{15}N$ values (average =

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+12.3‰ ± 2.5‰, $n = 13$) excluding one outlier, +4.9‰ (*F. vesiculosus*, 1997, Grassendale). Although the record is more stable, the $\delta^{15}N$ values are elevated in comparison to a background "natural" macroalgae range (+4‰ to +6‰). The herbaria $\delta^{15}N$ values are significantly more elevated than those in the 1821–1863 period (p value < 0.02, $n = 13$), which is interpreted as a result of wastewater treatment processes that elevate the ^{15}N content of sewage effluent. It is proposed that the stability of $\delta^{15}N$ in this time interval was a result of stricter controls on discharges, monitoring and increased chemical processing (*i.e.*, ammonia) in wastewater treatment plants.[25,61,72,73]

During this time interval, stricter regulations were implemented by the European Union on the UK government to address water quality issues. This legally enforced that the level of nitrate released into freshwater and marine environments be set to a maximum limit of 50 mg l^{-1} (or 11.3 mg l^{-1} of nitrate N).[65] Environmental action, regulations and investment around the Mersey Basin led to the reduction of BOD from 40 mg l^{-1} in the 1960s to ~7 mg l^{-1} in the River Mersey in the early 2000s;[34] the Mersey Estuary is still classified as eutrophic.

Mersey Estuary, 2022: a peak in the sewage era?

The macroalgae $\delta^{15}N$ data from September 2022 are very elevated (+16.5‰ ± 1.9‰) and significantly different from those of all previous time intervals, except for 1949–1983. $\delta^{15}N$ values are consistently more elevated with the lowest value at +12.8‰ and the highest value of +22.8‰.[34,69] Elevated $\delta^{15}N$ values >+16‰ are rarely recorded in macroalgae studies[45,74] (Gröcke *et al.*, unpub. data), and only a handful of studies have recorded values >20‰,[21,25,45,74] and none previously in the UK. Although BOD assessments indicate that the Mersey Estuary has improved, the Environment Agency reports that it is "not achieving good status" due to industry,[63] with no indication of water detriment from the water industry. The Environment Agency also gives "low confidence" that targets to reach "good" nitrogen levels will be achieved by 2027.[75,76] The herbaria $\delta^{15}N$ data indicate that the dominant 'nitrogen' pollution signal in the Mersey Estuary is caused by wastewater (*i.e.*, sewage) and not by nitrogen chemical pollutants (*i.e.*, artificial fertilisers). Overall, recent herbaria and modern macroalgae $\delta^{15}N$ datasets suggest that denitrified sewage input into the waters is extensive and well above the "natural" range expected for a coastal and estuarine environment in the North Atlantic.

Although there is a time-gap between the 1990−2013 and 2022 $\delta^{15}N$ datasets there is a clear and significant increase. Since 2012 the Environment Agency has been aware of illegal sewage discharges by United Utilities and has been accused of "knowingly permitting" such discharges to occur.[10] DEFRA states that CSOs are only permitted to discharge during periods of heavy, continuous rainfall and will be tightened to permit discharges only where "there is no adverse ecological impact" by 2050.[77] Although there is abundant social media evidence that water company regulations on discharging are not being adhered to,[78] additional evidence was presented in a recent BBC Panorama report documenting how pollution incidents are being covered up.[79]

The Liverpool sewerage infrastructure is one of the oldest sewerage systems in the UK,[80] and discharges predominantly through CSOs (*i.e.*, 84% of outlets to the River Mersey and Mersey Estuary). We interpret the elevated macroalgae $\delta^{15}N$ values as directly sourced from these CSOs which routinely discharge sewage effluent directly into the river. For example, in 2022 the River Mersey received a total of 3346 hours of sewage dumping (*i.e.*, 4.6 months)[43,81,82] dominantly occurring around Manchester and Warrington.[43,81–83] Although nitrate assimilation can lead to elevated residual nitrate $\delta^{15}N$ values, its impact is relatively minor in the order of 5‰ to 8‰.[84] Thus, denitrification processes must be a primary cause for generating elevated $\delta^{15}N$ recorded in macroalgae. There are four potential mechanisms and sources:

(1) denitrified sewage effluent from wastewater treatment plants. It is well documented that wastewater treatment processes that use anaerobic conditions can elevate effluent $\delta^{15}N$ values above +10‰;[85]

(2) increasing nitrogen productivity and deposition of nitrate as a consequence of elevated wastewater effluent discharges. Nitrogen consumption in the water profile *via* surface water productivity (*i.e.*, phytoplankton) will preferentially remove ^{14}N, resulting in more elevated nitrate $\delta^{15}N$ values in the water column. Burial of the nitrate and nitrogen-bound organic matter will also remove ^{14}N in preference to ^{15}N causing an increase in $\delta^{15}N$.

(3) denitrified groundwater nitrate. A 1999 study on groundwater nitrate indicated that sewage effluent was leaking into the aquifer beneath Liverpool (Whitehead *et al.* 1999) – this was based on nitrate $\delta^{15}N$ and microbiological analyses. However, the sample with the most elevated *E. coli* and faecal streptococci contents was not analysed for nitrate $\delta^{15}N$. The other samples in that study ranged between −11.9‰ and +13.2‰ (average = +6.8‰ ± 8.7‰), and therefore, it would seem that aquifer discharge is not a cause for elevated $\delta^{15}N$ in the Mersey Estuary. Further research is required to fully ascertain the effect of groundwater nitrate on this system; and

(4) denitrification in estuarine sediment. The tidal range for the Mersey Estuary is large (between 4 m to 10 m). Due to the volume of water being replenished daily it is reasonable to state that the water column would be well-oxygenated and, hence, increase the depth of the suboxic layer in the sediment. Nitrification of wastewater ammonia would occur in the water column, and nitrification in the sediment profile will produce elevated $\delta^{15}N$ pore-water ammonia values as nitrate is reduced in concentration.[86,87] Subsequent denitrification in the sediment profile is therefore a function of oxygen supply and penetration, as well as nitrate replenishment from the water column into the sediment profile. Although the Mersey Estuary has the largest total inorganic nitrogen load (3959×10^6 moles per year) in an estuary in the UK,[88] further research on nitrification and denitrification in Mersey Estuary sediment is required to understand its impact on the system.

Of the four options above, the most preferred explanation as a cause for the elevated $\delta^{15}N$ values in macroalgae in the Mersey Estuary is due to (1) and (2)—increased denitrified and raw

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wastewater sewage effluent causing high productivity and eutrophication in the River Mersey and Mersey Estuary. At present, water companies are undeterred by fines imposed by regulatory bodies[1] and have no incentive to change current operational practices (*i.e.*, releasing effluent during dry periods). Funding cuts to the Environment Agency have also limited their ability to properly monitor, respond, assess and impose fines on water companies in breach of environmental standards.[1,10,11] The decline in England river water quality (and subsequently, estuaries and coastal settings) is exacerbated by a continued, significant lack of investment from water companies to improve and repair their infrastructure to accompany increases in population and corresponding changes in present/future climate conditions; illegal discharging from CSOs has become so frequent as to become the 'norm'.[89] Although England water companies have until 2050 to achieve infrastructure upgrades and compliance, by that time the environmental damage to rivers, estuaries and coastal settings may be irreversible.[77] Continued nitrogen isotope monitoring of macroalgae will show whether improvements are made in the future or whether the Mersey Estuary will remain an environment impacted by sewage pollution in the 'sewage era'.

Conclusion

The Mersey Estuary has been identified as a heavily polluted environment since the early 1800s. Museum herbaria provide an excellent source of material to reconstruct historical pollution. Despite the number of available herbaria samples for this study, it has been possible to depict broadscale nitrogen isotope changes and trends in the Mersey Estuary since 1821.

A macroalgae herbaria $\delta^{15}N$ dataset from 1813 to 2013 reveals three major nitrogen pollution episodes: (1) 1821–1863 is dominated by raw sewage; (2) 1949–1983 is influenced by industrial, agricultural and treated sewage processes; and (3) 1990–2013 records treated and raw sewage pollution termed the 'sewage era'. Although BOD has decreased from very elevated levels in the 1960s, the River Mersey–Mersey Estuary still contains a significant proportion of nitrogen sewage pollution as interpreted using $\delta^{15}N$ macroalgae values. Privatisation of water companies in England in 1989 was driven by economics and not environmental sustainability. Its impact on herbaria $\delta^{15}N$ values was to stabilise the variability (and hence, the influence of varying nitrogen pollution sources). The modern macroalgae $\delta^{15}N$ record reflects increasing sewage nitrogen pollution into the River Mersey – through denitrified wastewater effluent and release of raw sewage. The very elevated $\delta^{15}N$ values are interpreted as a consequence of limited regulation, underinvestment, and legislation changes.

Poor water quality is an incessant problem in the UK—the elevated macroalgae $\delta^{15}N$ values recorded in both herbaria and modern samples from the Mersey Estuary highlight an ongoing failure to reduce nitrogen pollution in our hydrological environments. This study demonstrates the usefulness of macroalgae $\delta^{15}N$ in determining the nitrogen pollution source into the Mersey Estuary over the past 200 years and, hence, is indicative of a whole catchment source problem. The Mersey Estuary $\delta^{15}N$ record showcases how the nitrogen cycle has been severely influenced by human processes that have had a lasting impact on our riverine/estuarine environments. Large-scale nitrogen pollution from wastewater treatment plants is transforming the nitrogen cycle in UK rivers, estuaries and coastlines. This study highlights that the 'sewage era' is upon us and necessitates a critical shift in increased private investment into wastewater treatment infrastructure, stricter and immediate prosecution of policy breakers and a re-evaluation of environmental monitoring methods and policy.

Materials and methods

A total of 72 herbaria macroalgae specimens were sampled at the World Museum, Liverpool, providing a sample record from 1821 to 2018. The macroalgae specimens are from multiple locations and different macroalgae species for the Mersey Estuary and Liverpool South Docks (see Fig. 1); specific sample details are provided in the ESI.† A scalpel was used to remove the longest macroalgae thallus tips, which represent the last growth of the sample for all macroalgae specimens. The sample was then transferred into microcentrifuge tubes until processing for stable isotope analysis. It has been reported that nitrogen isotopic ratios are not impacted by paper type and so herbaria are assumed to be comparable to specimens pressed on modern acid-free paper.[32] Even with this knowledge we still inspected every herbarium sample to prove that no sample had herbarium paper attached. Herbarium specimens were compared to a modern geospatial sample set from the Mersey Estuary collected in September 2022. This sample set was collected for comparison with the herbaria samples – most of which were also collected in the summer season. Fifty *F. vesiculosus* and 22 *Ulva* sp. modern specimens were sampled from the Mersey Estuary in addition to 27 specimens collected from the South Docks (Fig. 1). Dock samples include 22 *Ulva* sp., nine *Cladophora* sp. and two C*allithamnion corymbosum*. Modern specimens were dried in an oven between 45 and 60 °C – this drying method has no impact on the bulk signature of macroalgae stable isotope ratios. Once dried, sub-samples of macroalgae and the most recent growing tip of *Fucus* sp. were weighed between 1.1 and 1.5 mg into tin capsules for stable isotope analysis. All nitrogen isotope analyses were carried out in the Stable Isotope Biogeochemistry Laboratory (SIBL) at Durham University. Nitrogen isotopes are reported against atmospheric nitrogen (AIR) and accuracy is continuously monitored with both in-house and international standards. In-house standards are calibrated against international standards (*e.g.*, IAEA-600, IAEA-N1, and IAEA-N2). Analytical uncertainty is typically $\pm 0.1‰$ (1 sd) for replicate analyses of our standards. Most herbaria specimens only had enough material for one analysis due to strict museum limitations on sample destruction. For further details on analytical methods see Bailes and Grocke[21] and Grocke *et al.*[21,26]

Conflicts of interest

The authors declare there are no conflicts of interest to declare.

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References

1 Environment Agency, *Water and sewerage company performance on pollution hits new low*, **https://www.gov.uk/government/news/water-and-sewerage-company-performance-on-pollution-hits-new-low**, accessed 4 March 2024.
2 J. Bevan, The state of our waters: the facts, **https://environmentagency.blog.gov.uk/2020/10/02/the-state-of-our-waters-the-facts/**, accessed 14 March 2023.
3 Environmental Audit Committee, *Water Quality in Rivers: Fourth Report of Session 2021–22*, London, 2022, **https://publications.parliament.uk/pa/cm5802/cmselect/cmenvaud/74/summary.html**, accessed 6 March 2024.
4 The Rivers Trust, *State of Our Rivers*, **https://theriverstrust.org/rivers-report-2024**, accessed 1 March 2024.
5 E. Gill, B. Horton, J. Gilbert, S. Riisnaes and E. Partridge, *Storm Overflow Evidence Project Final Report Prepared for: Water UK*, **https://assets.publishing.service.gov.uk/media/6182bad4e90e07197867ecd4/storm-overflows-evidence-project.pdf**, accessed 6 March 2024.
6 BBC, *Paul Whitehouse and Our Troubled Rivers*, **https://www.bbc.co.uk/programmes/m001jw74**, accessed 20 February 2024.
7 R. Bermejo, N. Golden, E. Schrofner, K. Knöller, O. Fenton, E. Serrão and L. Morrison, Biomass and nutrient dynamics of major green tides in Ireland: Implications for biomonitoring, *Mar. Pollut. Bull.*, 2022, **172**, 113318.
8 P. M. Glibert, Eutrophication, harmful algae and biodiversity—Challenging paradigms in a world of complex nutrient changes, *Mar. Pollut. Bull.*, 2017, **124**, 591–606.
9 V. H. Smith, Eutrophication of Freshwater and Coastal Marine Ecosystems A Global Problem, *Environ. Sci. Pollut. Res.*, 2003, **10**, 126–139.
10 House of Commons, Oral Evidence: Work of the Environment Agency, HC 221, *Environment, Food and Rural Affairs Committee*, **https://committees.parliament.uk/oralevidence/12822/html/**, 2023.
11 S. Lovett, Environment Agency funding cut by 50% over past decade as sewage spills rise, analysis shows, The Independent, **https://www.independent.co.uk/climate-change/news/water-pollution-sewage-environment-agency-funding-b2154848.html#**, 2022, accessed 2 March 2024.
12 H. Briggs, Citizen scientists join fight to clean up rivers, *BBC Science & Environment*, **https://www.bbc.co.uk/news/science-environment-63747838**, 2022, accessed 4 March 2024.
13 J. Woodward, To clean up England's rivers we need to know how much sewage is dumped – but water firms won't tell us, *University of Manchester*, **https://www.manchester.ac.uk/discover/news/sewage-water-firms-wont-tell-us/**, accessed 4 March 2024.
14 J. Tucker, N. Sheats, A. E. Giblin, C. S. Hopkinson and J. P. Montoya, Using stable isotopes to trace sewage-derived material through Boston Harbor and Massachusetts Bay, *Mar. Environ. Res.*, 1999, **48**, 353–375.
15 S. D. Costanzo, M. J. Oõdonohue, W. C. Dennison, N. R. Loneragaǹ and M. Thomas, A New Approach for Detecting and Mapping Sewage Impacts, *Mar. Pollut. Bull.*, 2001, **42**, 149–156.
16 C. Savage and R. Elmgren, Macroalgal (*Fucus vesiculosus*) $\delta^{15}N$ values trace decrease in sewage influence, *Ecol. Appl.*, 2004, **14**, 517–526.
17 J. Samper-Villarreal, Strengths and challenges of $\delta^{15}N$ to identify anthropogenic nutrient loading in coastal systems, *Isot. Environ. Health Stud.*, 2020, **56**, 700–712.
18 R. García-Seoane, J. A. Fernández, R. Villares and J. R. Aboal, Use of macroalgae to biomonitor pollutants in coastal waters: Optimization of the methodology, *Ecol. Indicat.*, 2018, **84**, 710–726.
19 R. Cohen and P. Fong, Experimental evidence supports the use of $\delta^{15}N$ content of the opportunistic green macroalga Enteromorpha intestinalis to determine nitrogen sources to estuaries, *J. Phycol.*, 2005, **41**, 229–448.
20 D. Xue, J. Botte, B. De Baets, F. Accoe, A. Nestler, P. Taylor, O. Van Cleemput, M. Berglund and P. Boeckx, Present limitations and future prospects of stable isotope methods for nitrate source identification in surface- and groundwater, *Water Res.*, 2009, **43**, 1159–1170.
21 I. R. Bailes and D. R. Gröcke, Isotopically labelled macroalgae: A new method for determining sources of excess nitrogen pollution, *Rapid Commun. Mass Spectrom.*, 2020, **34**, e8951.
22 L. Orlandi, E. Calizza, G. Careddu, P. Carlino, M. L. Costantini and L. Rossi, The effects of nitrogen pollutants on the isotopic signal ($\Delta^{15}N$) of *Ulva lactuca*: Microcosm experiments, *Mar. Pollut. Bull.*, 2017, **115**, 429–435.
23 A. S. Bateman and S. D. Kelly, Fertilizer nitrogen isotope signatures, *Isot. Environ. Health Stud.*, 2007, **43**, 237–247.
24 T. H. E. Heaton, Isotopic studies of nitrogen pollution in the hydrosphere and atmosphere: A review, *Chem. Geol. (Isotope Geoscience Section)*, 1986, **59**, 87–102.
25 F. C. Alldred, D. R. Gröcke, C. Y. Leung, L. P. Wright and N. Banfield, Diffuse and concentrated nitrogen sewage pollution in island environments with differing treatment systems, *Sci. Rep.*, 2023, **13**, 4838.
26 D. R. Gröcke, B. Racionero-Gómez, J. W. Marschalek and H. C. Greenwell, Translocation of isotopically distinct

Open Access Article. Published on 29 2024. Downloaded on 8/8/2025 5:51:10 AM. This article is licensed under a Creative Commons Attribution 3.0 Unported Licence.

macroalgae: A route to low-cost biomonitoring?, *Chemosphere*, 2017, **184**, 1175–1185.
27 G. H. Pyke and P. R. Ehrlich, Biological collections and ecological/environmental research: A review, some observations and a look to the future, *Biol. Rev.*, 2010, **85**, 247–266.
28 A. M. Lister, S. J. Brooks, P. B. Fenberg, A. G. Glover, K. E. James, K. G. Johnson, E. Michel, B. Okamura, M. Spencer, J. R. Stewart, J. A. Todd, E. Valsami-Jones and J. Young, *Trends Ecol. Evol.*, 2011, **26**, 153–154.
29 C. Lavoie, Biological collections in an ever changing world: Herbaria as tools for biogeographical and environmental studies, *Perspect. Plant Ecol. Evol. Systemat.*, 2013, **15**, 68–76.
30 M. R. Pie, S. P. Batke, J. Reyes-Chávez and T. Dallimore, Fern and lycophyte niche displacement under predicted climate change in Honduras, *Plant Ecol.*, 2022, **223**, 613–625.
31 C. C. Davis, The herbarium of the future, *Trends Ecol. Evol.*, 2022, **38**, 412–423.
32 E. A. Miller, S. E. Lisin, C. M. Smith and K. S. Van Houtan, Herbaria macroalgae as a proxy for historical upwelling trends in Central California, *Proc. R. Soc. B*, 2020, **287**, 20200732.
33 B. Alfonso, M. Sansón, C. Sangil, F. J. Expósito, J. P. Díaz and J. C. Hernández, Herbarium macroalgae specimens reveal a rapid reduction of thallus size and reproductive effort related with climate change, *Mar. Environ. Res.*, 2021, **174**, 105546.
34 S. J. Hawkins, K. A. O'Shaughnessy, L. A. Adams, W. J. Langston, S. Bray, J. R. Allen, S. Wilkinson, K. Bohn, N. Mieszkowska and L. B. Firth, Recovery of an urbanised estuary: Clean-up, de-industrialisation and restoration of redundant dock-basins in the Mersey, *Mar. Pollut. Bull.*, 2020, **156**, 111150.
35 Mersey Basin Campaign, *River Mersey*, **https://www.merseybasin.org.uk/#:~:text=TheMerseyBasinCampaignbegan,MerseyBasinCampaignin2010**.
36 N. Ritchie-Noakes, *Liverpool's Historic Waterfront: The World's First Mercantile Dock System*, H.M. Stationary Office, London, 1984.
37 ONS, Census 2021, **https://www.ons.gov.uk/visualisations/censuspopulationchange/E08000003/**, accessed 7 October 2023.
38 L. R. Burton, The Mersey Basin: An historical assessment of water quality from an anecdotal perspective, *Sci. Total Environ.*, 2003, **314–316**, 53–66.
39 L. R. Burton, A. Howard and B. Goodall, Construction of a historical water pollution index for the Mersey Basin, *Area*, 2003, **35**, 438–448.
40 P. G. W. Jones and S. M. Haq, The Distribution of *Phaeocystis* in the Eastern Irish Sea, *J. Mar. Sci.*, 1963, **28**, 8–20.
41 S. Meredith, Water Privatisation: the dangers and the benefits, *Long. Range Plan.*, 1992, **25**, 72–81.
42 BBC, River Mersey should be sewage free by 2030, says metro mayor, 2021, **https://www.bbc.co.uk/news/uk-england-merseyside-59406932**, last accessed 3 March 2024.
43 The Rivers Trust, Sewage in Our Rivers, 2024, **https://theriverstrust.org/sewage-map**, accessed 7 March 2024.
44 B. Jessop, Neoliberalization, uneven development, and Brexit: further reflections on the organic crisis of the British state and society, *Eur. Plann. Stud.*, 2018, **26**, 1728–1746.
45 M. L. Dailer, R. S. Knox, J. E. Smith, M. Napier and C. M. Smith, Using $\delta^{15}N$ values in algal tissue to map locations and potential sources of anthropogenic nutrient inputs on the island of Maui, Hawai'i, USA, *Mar. Pollut. Bull.*, 2010, **60**, 655–671.
46 J. R. Allen, S. B. Wilkinson and S. J. Hawkins, Redeveloped docks as artificial lagoons: The development of brackish-water communities and potential for conservation of lagoonal species, *Aqua. Conserv.: Mar. Fresh. Ecosyst.*, 1995, **5**, 299–309.
47 S. B. Wilkinson, W. Zheng, J. R. Allen, N. J. Fielding, V. C. Wanstall, G. Russell and S. J. Hawkins, Water quality improvements in Liverpool docks: The role of filter feeders in algal and nutrient dynamics, *Mar. Ecol.*, 1996, **17**, 197–211.
48 R. M. Connolly, D. Gorman, J. S. Hindell, T. N. Kildea and T. A. Schlacher, High congruence of isotope sewage signals in multiple marine taxa, *Mar. Pollut. Bull.*, 2013, **71**, 152–158.
49 R. J. Pruell, B. K. Taplin, A. J. Oczkowski, J. S. Grear, W. G. Mendoza, A. R. Pimenta, A. R. Hanson and K. M. Miller, Nitrogen isotope fractionation in a continuous culture system containing phytoplankton and blue mussels, *Mar. Pollut. Bull.*, 2022, **150**, 110745.
50 I. G. Viana and A. Bode, Stable nitrogen isotopes in coastal macroalgae: Geographic and anthropogenic variability, *Sci. Total Environ.*, 2013, **443**, 887–895.
51 D. M. Sigman, M. A. Altabet, D. C. McCorkle, R. Francois and G. Fischer, The $\delta^{15}N$ of nitrate in the Southern Ocean: Nitrogen cycling and circulation in the ocean interior, *J. Geophys. Res.: Oceans*, 2000, **105**, 19599–19614.
52 J. Galloway, A. Leach, J. W. Erisman and A. Bleeker, Nitrogen: the historical progression from ignorance to knowledge, with a view to future solutions, *Soil Res.*, 2017, **55**, 417–424.
53 P. Cao, C. Lu and Z. Yu, Historical nitrogen fertilizer use in agricultural ecosystems of the contiguous United Stages during 1850-2015: application rate, timing and fertiliser types, *Earth Syst. Sci. Data*, 2018, **10**, 969–984.
54 L. Rosenthal, *The River Pollution Dilemma in Victorian England: Nuisance Law versus Economic Efficiency*, Routledge, London, 1st edn, 2014.
55 M. Hughes, The Victorian London sanitation projects and the sanitation of projects, *Int. J. Proj. Manag.*, 2013, **31**, 682–691.
56 E. Porter, *Pollution in Four Industrialised Estuaries: Studies in Relation to Changes in Population and Industrial Development; Four Case Studies Undertaken for the Royal Commission on Environmental Pollution*, H.M. Stationary Office, London, 1973, **https://archive.org/details/pollutioninfouri0000port**.
57 R. J. Davenport, M. Satchell and L. M. W. Shaw-Taylor, Cholera as a 'sanitary test' of British cities, 1831–1866, *Hist. Fam.*, 2019, **24**, 404–438.
58 S. Halliday, *The Great Stink of London: Sir Joseph Bazalgette and the Cleansing of the Victorian Metropolis*, Sutton Publishing, 1999.

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59 H. S. Lee and B. Liao, Anaerobic membrane bioreactors for wastewater treatment: Challenges and opportunities, *Water Environ. Res.*, 2021, **93**, 993–1004.
60 R. E. B. Reid, B. E. Crowley and R. J. Haupt, The prospects of poop: a review of past achievements and future possibilities in faecal isotope analysis, *Biol. Rev.*, 2023, **98**, 2091–2113.
61 M. I. Bird, J. Haig, S. Ulm and C. Wurster, A carbon and nitrogen isotope perspective on ancient human diet in the British Isles, *J. Archaeol. Sci.*, 2022, **137**, 105516.
62 M. J. Whelan, C. Linstead, F. Worrall, S. J. Ormerod, I. Durance, A. C. Johnson, D. Johnson, M. Owen, E. Wiik, N. J. K. Howden, T. P. Burt, A. Boxall, C. D. Brown, D. M. Oliver and D. Tickner, Is water quality in British rivers "better than at any time since the end of the Industrial Revolution", *Sci. Total Environ.*, 2022, **843**, 157014.
63 N. J. K. Howden, T. P. Burt, F. Worrall, M. J. Whelan and M. Bieroza, Nitrate concentrations and fluxes in the River Thames over 140 years (1868–2008): Are increases irreversible?, *Hydrol. Processes*, 2010, **24**, 2657–2662.
64 P. D. Jones, The Mersey Estuary - Back from the dead? Solving a 150-Year Old Problem, *Water Environ. J.*, 2000, **14**, 124–130.
65 OFWAT, *The Development of the Water Industry in England and Wales*, 2006, **https://www.ofwat.gov.uk/publication/the-development-of-the-water-industry-in-england-and-wales/**.
66 K. M. Rogers, Stable carbon and nitrogen isotope signatures indicate recovery of marine biota from sewage pollution at Moa Point, New Zealand, *Mar. Pollut. Bull.*, 2003, **46**, 821–827.
67 UK Government, *Control of Pollution Act 1974*, UK Government, London, 1974, **https://www.legislation.gov.uk/ukpga/1974/40**.
68 W. Howarth, Water Pollution: Improving the Legal Controls, *J. Environ. Law*, 1989, **1**, 25–37.
69 S. Kidd and D. Shaw, The Mersey Basin and its River Valley Initiatives: An appropriate model for the management of rivers?, *Local Environ.*, 2000, **5**, 191–209.
70 UK Government, *The Water Act*, UK Government, 1989, **https://www.legislation.gov.uk/ukpga/1989/15/contents**.
71 A. Schaefer, Corporate greening and changing regulatory regimes: The UK water industry, *Bus. Strat. Environ.*, 2009, **18**, 320–333.
72 DEFRA, *Sewage Treatment in the UK: UK Implementation of the EC Urban Waste Water Treatment Directive*, London, 2002, **https://www.gov.uk/government/publications/sewage-treatment-in-the-uk-2002**.
73 K. Bakker, Neoliberalizing nature? market environmentalism in water supply in England and Wales, *Ann. Assoc. Am. Geogr.*, 2005, **95**, 542–565.
74 S. Van Wynsberge, F. Antypas, M. Brisset, A. Desnues, L. Jamet, L. Lagourgue, C. Payri, T. Jauffrais and H. Lemonnier, A new set of N isotopic reference values for monitoring *Ulva* green tides in coral reef ecosystems, *Mar. Pollut. Bull.*, 2024, **200**, 116152.
75 G. Aertebjerg, J. Carstensen, K. Dahl, J. Hansen, K. Nygaard, B. Rygg, K. Sørensen, G. Severinsen, S. Casartelli, W. Schrimpf, C. Schiller and J. N. Druon, *Eutrophication in Europe's Coastal Waters*, Copenhagen, 2001.
76 Environment Agency, Mersey Water Body, **https://environment.data.gov.uk/catchment-planning/WaterBody/GB531206908100**, accessed 3 March 2024.
77 DEFRA, *Consultation on the Government's Storm Overflows Discharge Reduction Plan*, 2022, **https://assets.publishing.service.gov.uk/media/65115ef02f404b000dc3d823/Government_response_to_the_overflows_consultation.pdf**, last accessed, 6 March 2024.
78 J. Hobson and B. Holmes, *BBC Online*, 2023, **https://www.bbc.co.uk/news/uk-england-merseyside-66455794#**, last accessed 3 March 2024.
79 BBC Panorama, The Water Pollution Cover-Up, 2023, **https://www.bbc.co.uk/programmes/m001t4g5**, last accessed 6 March 2024.
80 National Rivers Authority, *The Mersey Estuary: a Report on Environmental Quality*, Harlequin Colourprint, Bristol, 1995.
81 United Utilities, *Storms Overflow Report*, Warrington, 2022, **https://www.unitedutilities.com/corporate/responsibility/environment/**, last accessed 7 April 2023.
82 L. Thorp, More than 147 days' worth of raw sewage dumped in Merseyside water last year, Liverpool Echo, 2023, **https://www.liverpoolecho.co.uk/news/liverpool-news/more-147-days-worth-raw-26698627#**, last accessed 6 March 2024.
83 Top of the Poops, Pollution Summary, 2022, **https://top-of-the-poops.org/waterway/united-utilities/river-mersey**, accessed 6 March 2024.
84 F. Fripiat, A. Martinez-Garcia, S. Fawcett, P. Kemeny, A. Studer, S. Smart, F. Rubach, S. Oleynik, D. Sigman and G. Haug, The isotope effect of nitrate assimilation in the Antarctic Zone: Improved estimates and paleoceanographic implications, *Geochim. Cosmochim. Acta*, 2019, **247**, 261–279.
85 T. Onodera, K. Komatsu, A. Kohzu, G. Kanaya, M. Mizuochi and K. Syutsubo, Evaluation of stable isotope ratios ($\delta^{15}N$ and $\delta^{18}O$) of nitrate in advanced sewage treatment processes: Isotopic signature in four process types, *Sci. Total Environ.*, 2021, **762**, 144120.
86 M. F. Lehmann, D. M. Sigman, D. C. McCorkle, J. Granger, S. Hoffmann, G. Cane and B. G. Brunelle, The distribution of nitrate $^{15}N/^{14}N$ in marine sediments and the impact of benthic nitrogen loss on the isotopic composition of oceanic nitrate, *Geochim. Cosmochim. Acta*, 2007, **71**, 5384–5404.
87 M. Alkhatib, M. F. Lehmann and P. A. del Giorgio, The Nitrogen Isotope Effect of Benthic Remineralisation-Nitrification-Denitrification Coupling in an Estuarine Environment, *Biogeosciences*, 2012, **9**, 1633–1646.
88 D. B. Nedwell, L. F. Dong, A. Sage and G. J. C. Underwood, Variations of the Nutrients Loads to the Mainland U.K. Estuaries: Correlation with Catchment Areas, Urbanisation and Coastal Eutrophication, *Estuar. Coast Shelf Sci.*, 2002, **54**, 951–970.
89 T. Giakoumis and N. Voulvoulis, Combined sewer overflows: relating event duration monitoring data to wastewater systems' capacity in England, *Environ. Sci. (Camb.)*, 2023, **9**, 707–722.