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Research Article

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Selective Removal of Organoarsenic Species and Aromatic Amine Compounds from Industrial Wastewater Using a Periwinkle Shell-Derived Chitosan–Lemon Waste Biofunctionalized Composite

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Abstract

Organoarsenic species and aromatic amines are toxicologically significant industrial wastewater pollutants that have been comparatively neglected as chitosan-graft target contaminants relative to dyes, heavy metals, and phthalates. This study valorised two co-located agro-industrial waste streams, periwinkle shell and lemon fruit waste, into a chitosan–lemon waste graft (CLWG) and evaluated its performance against real textile-associated industrial effluent from Kaduna, Nigeria. Chitosan was extracted from periwinkle shells and grafted with citric-acid-treated lemon waste; unmodified chitosan and the CLWG were characterised by scanning electron microscopy with energy-dispersive X-ray spectroscopy (SEM-EDX) and attenuated total reflectance Fourier-transform infrared (ATR-FTIR) spectroscopy, and applied in batch trials at 0.5 g and 1.0 g doses over 4 h and 8 h contact times. SEM revealed a marked shift from the compact, fine-grained morphology of unmodified chitosan to a coarser, more heterogeneous, and porous surface after grafting, and EDX confirmed successful modification through the emergence of nitrogen (0.00 to 4.65 atomic %) alongside a sharp decline in calcium (78.22 to 2.21 atomic %), consistent with exposure of the chitosan-derived phase and leaching of the native shell mineral during synthesis. ATR-FTIR showed new carbonyl and altered glycosidic bands after grafting, and further band splitting and new low-frequency features after adsorption, indicating direct functional-group involvement in pollutant binding. Gas chromatography-mass spectrometry identified an aromatic amine (benzenamine, 2,6-diethyl-), a nitrosated aniline derivative, and an arsenous acid trimethylsilyl ester in the untreated effluent; all three were undetectable at every tested CLWG dose and contact time, while two transient nitrogen-containing aromatic byproducts observed only at the lowest dose and shortest contact time were themselves absent by 8 h or at the 1 g dose. These findings indicate that a low-cost, dual-agro-waste-valorised chitosan graft can achieve apparent complete removal of both organoarsenic and aromatic amine pollutant classes from real industrial wastewater, supporting its potential as a circular-economy remediation strategy for resource-limited settings.

Keywords: Chitosan Grafting; Periwinkle Shell Valorization; Organoarsenic Removal; Aromatic Amine Adsorption; SEM-EDX Characterization; ATR-FTIR; GC-MS Wastewater Profiling

1. Introduction

Industrial wastewater discharged into receiving water bodies frequently carries trace to ultratrace concentrations of arsenic-bearing species, among the most extensively documented environmental toxicants worldwide. The World Health Organization and the International Agency for Research on Cancer (IARC) classify arsenic and inorganic arsenic compounds as carcinogenic to humans, with long-term exposure first manifesting as skin pigmentation changes and hyperkeratosis that may progress to skin cancer, and with established associations with bladder and lung cancer [3]. A systematic review of the global epidemiological evidence further reported a consistent dose-response relationship between drinking-water arsenic and skin lesions even at exposure levels close to the WHO guideline value of 10 $\mu\text{g/L}$, indicating that risk persists well below historically "high-exposure" thresholds [1]. Mechanistically, the trivalent arsenic species typified by arsenous acid (arsenite) reacts readily with the sulfhydryl ($-\text{SH}$) groups of proteins, inhibiting key enzymes, disrupting oxidative metabolism, and producing morphological damage to liver mitochondria [2]. Because industrial effluent is a major anthropogenic pathway for arsenic entry into surface and groundwater, and because trivalent inorganic arsenic is markedly more toxic and mobile than its methylated organic counterparts, the recurrent detection of an arsenite-derived organoarsenic species in raw industrial wastewater represents a quantifiable and non-trivial human and ecological health hazard, providing strong justification for its targeted removal prior to discharge [2,3].

A second pollutant class of comparable concern is the aromatic amines, industrial chemicals widely used as intermediates in the manufacture of dyes, pesticides, pharmaceuticals, and pigments. Epidemiological reviews attribute up to 25% of occupational bladder cancer cases in some Western industrial regions to aromatic amine exposure, with several arylamines formally classified by IARC as proven or probable human carcinogens [4]. Beyond carcinogenicity, aromatic amines are potent inducers of methaemoglobinaemia, a form of chemically induced oxygen starvation, and are considered highly toxic due to their mutagenic activity in a range of test systems [5]. Critically for wastewater-treatment research, aromatic amines are not merely a human-exposure concern but are directly implicated in the genotoxic character of industrial effluent itself: a recent environmental review identified aromatic amines as partially responsible for the mutagenic and genotoxic traits of industrial wastewater, noting that insufficiently treated effluent contributes measurably to the mutagenic burden of receiving surface waters [6]. Given that the effluent examined in the present study originates from an industrial area associated with textile and allied processing, where azo-dye chemistry is a plausible source of aromatic amine residues, the presence of an aromatic amine in the untreated wastewater is consistent with known industrial sources and reinforces the case for its deliberate removal. More broadly, comprehensive characterisation of the full range of emerging organic and inorganic contaminants in industrial wastewater remains analytically challenging, particularly for trace-level species that fall outside routine regulatory monitoring panels, underscoring the value of untargeted GC-MS profiling of the type applied in this study [7].

Chitosan, the deacetylated derivative of chitin, is conventionally sourced from crustacean shell waste, but shell-bearing gastropods such as the periwinkle offer an underexploited, low-cost regional

alternative, particularly in coastal and riverine West African communities where periwinkle shells accumulate as a market and food-processing waste stream. Gbenebor *et al.* [8] established a reproducible sequential demineralisation–deproteinisation–deacetylation route for extracting chitin and chitosan from periwinkle and snail shells, reporting a rough, mineral-reinforced surface morphology attributable to the shell's native calcium carbonate architecture, a finding corroborated by subsequent periwinkle-specific extraction studies. Onosakponome *et al.* [9] extracted chitin and chitosan from *Tympanotonos fuscatus* periwinkle shells under varying decolourisation conditions, reporting chitosan yields of 28.0–48.1% and degrees of deacetylation of 46.3–63.5%, with FTIR spectra confirming the expected O–H, N–H, and glycosidic functional signatures of chitosan. In a comparative seafood-waste study, Ewii *et al.* [10] extracted and characterised chitosan from crab, periwinkle, and snail shells side by side, reporting a periwinkle-derived chitosan yield of 16.3% with a degree of deacetylation above 50% and the highest ash content among the three sources (10.5%), reflecting incomplete removal of the shell's mineral phase, a pattern also seen in other biogenic chitosan sources such as horse mussel shell [11]. Complementing these extraction studies, Andrew *et al.* [12] reviewed the use of scanning electron microscopy for morphological and microanalytical characterisation of chitosan-based nanocomposites, underscoring SEM-EDX as the standard technique for confirming surface modification and elemental change following chitosan grafting or compositing, an approach adopted directly in the characterisation strategy of the present study. This pattern is reinforced by the broader modification literature: Bhatnagar and Sillanpää [13] catalogued the wide range of chitin- and chitosan-based materials applied to water and wastewater detoxification, Crini [14] established chitosan and related biopolymers as viable low-cost alternatives to activated carbon for pollutant removal, and more recent syntheses by Kyzas and Bikiaris [15], Saheed *et al.* [16], and Atangana *et al.* [17] confirm that chemical and physical modification of chitosan, including grafting, crosslinking, and compositing, is now a mainstream strategy for tailoring adsorptive selectivity. None of these reviews, however, identify organoarsenic species as a well-represented chitosan-graft target class, and only a minority address aromatic amines directly, reinforcing the gap this study addresses. Wan Ngah *et al.* [18] similarly reviewed chitosan composites for dye and heavy-metal adsorption, further illustrating the dye/metal/phthalate skew in the existing pollutant-class coverage that the present focus on organoarsenic and aromatic amine removal seeks to redress.

Beyond its extraction, chitosan's native amine and hydroxyl functional groups make it an intrinsically attractive adsorbent, and grafting or compositing chitosan with secondary materials has repeatedly been shown to enhance its adsorptive performance, stability, and reusability. Fan *et al.* [19] grafted magnetic chitosan with graphene oxide to enhance methylene blue dye uptake while enabling magnetic solid–liquid separation, and Okoya *et al.* [20] demonstrated that chitosan-coated carbon achieved favourable Langmuir/Freundlich equilibrium for chromium (VI) and lead(II) removal from industrial effluent. Directly relevant to the pollutant classes targeted here, Salim *et al.* [21] reported the comparative adsorption of phthalate esters on chitosan beads via hydrogen-bonding and complexation mechanisms – phthalates being ubiquitous, endocrine-disrupting plasticiser residues whose occurrence, environmental fate, and health significance have been extensively reviewed elsewhere [22] – while Vafakish and Wilson [23] synthesised an aniline-grafted chitosan adsorbent explicitly for aromatic

amine and metal-ion capture, demonstrating that amine-functionalised chitosan grafts can selectively target aromatic nitrogenous pollutants of the type examined in this work. Other recent grafted-chitosan systems have targeted a broad range of emerging organic contaminants: Tzereme *et al.* [24] grafted chitosan with itaconic acid for diclofenac removal, Gubitosa *et al.* [25] developed chitosan–TiO₂ composite films for 2,4-dinitrophenol removal, Dehghani *et al.* [26] immobilised nanoscale zero-valent iron on chitosan for bisphenol A removal, and Monir *et al.* [27] most recently reported a novel grafted bio-based chitosan adsorbent for selective multi-pollutant removal from real wastewater. Collectively, this body of work establishes grafting as a validated strategy for enhancing chitosan's adsorptive selectivity and capacity, but also reveals that the great majority of grafted-chitosan studies have targeted dyes, heavy metals, phthalates, or bisphenol A, leaving organoarsenic species and aromatic amines, despite their comparable or greater toxicological significance, comparatively unexamined as chitosan-graft target pollutants in real industrial effluent.

The industrial wastewater matrix examined in this study is representative of a broader Nigerian treatment gap. Yusuff and Sonibare [28] characterised textile-industry effluent in Kaduna and reported that discharges routinely exceeded national and international physicochemical and pollutant-load standards, a finding consistent with the untreated effluent profiled here. Comparable treatment shortfalls have been documented elsewhere in the country's industrial base, including Niger Delta oil-refinery wastewater [29], decentralised food- and beverage-processing effluent [30], and pesticide- and heavy-metal-laden industrial discharge remediated using nanoparticle-based approaches [31]. Collectively, these studies indicate that low-cost, locally sourced remediation materials such as the CLWG developed here are of direct practical relevance to Nigeria's industrial wastewater management needs, and that the treatment gap they address is neither isolated to Kaduna nor to the textile sector specifically.

The literature reviewed above establishes three converging points: first, that organoarsenic species and aromatic amines are pollutants of serious, well-documented human and environmental health concern; second, that periwinkle shell waste is a viable, low-cost, regionally abundant source of chitosan; and third, that grafting chitosan with a secondary biobased material reliably improves its adsorptive performance, yet has rarely been applied specifically to organoarsenic or aromatic amine removal from real industrial effluent. This study addresses that gap by valorising two co-located agro-industrial waste streams, periwinkle shell and lemon fruit waste, into a single low-cost, biofunctionalized adsorbent, and by evaluating its performance against real, uncharacterised industrial wastewater rather than a synthetic single-pollutant solution. The aim of this study is therefore to synthesise and characterise a chitosan–lemon waste graft (CLWG) and to evaluate its efficiency for the removal of organoarsenic species, aromatic amine, and other co-occurring organic pollutants from real industrial wastewater. The specific objectives are to: (i) extract chitosan from periwinkle shell waste and synthesise a chitosan–lemon waste graft using citric acid as a cross-linking agent; (ii) characterise the unmodified chitosan and the resulting graft using scanning electron microscopy with energy-dispersive X-ray spectroscopy (SEM-EDX) and attenuated total reflectance Fourier-transform infrared (ATR-FTIR) spectroscopy to confirm successful grafting; (iii) apply the graft in batch adsorption trials against real industrial effluent

at two dosages (0.5 g and 1.0 g) and two contact times (4 h and 8 h); and (iv) profile the treated and untreated wastewater by gas chromatography–mass spectrometry (GC-MS) to quantify the removal of organoarsenic species, aromatic amine, and other major organic pollutant classes.

2. Materials and Methods

2.1 Materials, Reagents, and Study Area

Periwinkle shells and lemon fruit waste were sourced from open markets and food-processing sites within Kaduna metropolis, Nigeria. Analytical-grade hydrochloric acid, sodium hydroxide, acetic acid, citric acid, and dichloromethane were procured from licensed chemical vendors in Kaduna North Local Government Area. Industrial (textile and allied) effluent was collected from the Makera Industrial Area, Kakuri, Kaduna State, and used as the real wastewater matrix throughout the study. Instrumentation comprised a scanning electron microscope with energy-dispersive X-ray spectroscopy (SEM-EDX), an attenuated total reflectance Fourier-transform infrared (ATR-FTIR) spectrometer, an Agilent 19091S-433UI gas chromatograph–mass spectrometer (GC-MS), a laboratory oven, and a muffle furnace.

2.2 Extraction of Chitosan from Periwinkle Shell Waste

Chitosan was extracted following the sequential demineralisation–deproteinisation–deacetylation route of Gbenedor et al. [8], adapted as follows. Raw periwinkle shells were washed, briefly boiled to remove residual tissue, oven-dried (60 °C), and milled to a uniform powder. Demineralisation was performed with 2 M HCl (1:10 w/v, room temperature, 1–3 h) until effervescence ceased, followed by washing to neutral pH. Deproteinisation used 2 M NaOH (1:10 w/v, 65–90 °C, 1–4 h), with an optional 30% H₂O₂ bleaching step (1–3 h). Deacetylation of the dried chitin was carried out with 40% (w/v) NaOH (1:10 w/v, 100–120 °C, 4 h, with fresh NaOH introduced partway through), and the product was washed to neutral pH, ethanol-rinsed, oven-dried (50–60 °C, 12–24 h), and milled to a fine powder for use in grafting.

2.3 Preparation of Lemon Waste and Synthesis of the Chitosan–Lemon Waste Graft (CLWG)

Lemon fruit waste was washed, oven-dried (60 °C, 24 h), milled, and split into two equal portions: one carbonised to ~80% char and alkali-treated (0.1 M NaOH, 60–70 °C, 30 min), the other alkali-treated directly under the same conditions without carbonisation. The two portions were recombined in a 1:1 ratio to give the pre-treated lemon waste precursor. For grafting, 2 g of pre-treated lemon waste was soaked in citric acid solution (1 g/10 mL, 30 min) while 1 g of periwinkle-derived chitosan was separately dissolved in 2% (v/v) HCl; the citric acid–lemon waste mixture was then added slowly to the dissolved chitosan and reacted at 50–60 °C for 60 min under gentle stirring. The pH of the reacted mixture was raised to 8.5 with NaOH to precipitate the graft, which was allowed to settle overnight, cured in an oven (100–120 °C) to ~30% carbonisation, and pulverised to give the chitosan–lemon waste graft (CLWG) used in all subsequent experiments.

2.4 Batch Adsorption Experiments

Batch adsorption trials were conducted in duplicate under four dose/contact-time combinations (0.5g/4h, 0.5g/8h, 1g/4h and 1g/8h), using 100 cm³ of industrial wastewater per run at ambient temperature (27 °C) and pH 7.2 with occasional stirring. After each run, the filtrate was reserved for GC-MS profiling, and the spent solid from the highest-dose, longest-contact condition (1.0 g, 8 h) was dried and reserved for ATR-FTIR analysis of the post-adsorption surface.

2.5 Characterisation: SEM-EDX and ATR-FTIR

Unmodified chitosan and the CLWG were mounted on carbon adhesive stubs, sputter-coated, and examined by scanning electron microscopy in secondary-electron mode (15 kV, backscattered-detector full mode, 883 µm field of view) to compare surface morphology before and after grafting, with coupled energy-dispersive X-ray spectroscopy (EDX) used to obtain elemental composition and confirm the appearance of new elements as evidence of successful modification, following the general approach of Goldstein *et al.* [32], Egerton [33], and Andrew *et al.* [12]. Unmodified chitosan, the CLWG, and the dried spent adsorbent from the 1.0 g/8 h batch were each analysed by ATR-FTIR over 4000–400 cm⁻¹ to identify characteristic functional groups (O–H, N–H, C=O, C–O–C) and to track band shifts, new bands, or intensity changes as evidence of grafting and subsequent pollutant uptake [11].

2.6 Sample Extraction and GC-MS Analysis

Target organic pollutants were isolated from each wastewater sample (untreated and the four treated filtrates) by liquid–liquid extraction: a 500 mL aliquot was partitioned three times against dichloromethane/hexane in a separatory funnel, and the combined organic extracts were dried over anhydrous sodium sulphate, concentrated to dryness under nitrogen, and reconstituted in hexane/acetone prior to analysis. Extracts were analysed on an Agilent 19091S-433UI GC-MS fitted with an HP-5ms Ultra Inert capillary column (30 m × 250 µm × 0.25 µm), using helium carrier gas (0.974 mL/min, 7.36 psi head pressure), a 250 °C injector temperature, a 1 µL injection volume, and an oven programme spanning 0–325 °C (350 °C operating maximum). Compounds were identified against the instrument spectral library by retention time and CAS registry number, consistent with standard automated electron-impact mass-spectral library-matching approaches for this instrument class [34], and relative abundance was expressed as chromatographic peak area percentage, allowing direct comparison of pollutant profiles, including organoarsenic and aromatic amine species, between the untreated and treated wastewater samples across all four dose/contact-time conditions.

2.7 Data Treatment and Uncertainty

Batch adsorption trials were performed in duplicate for each dose/contact-time combination (Section 2.4). SEM-EDX and ATR-FTIR characterisation was performed in point-analysis/single-spectrum mode for each sample, and GC-MS profiling reflects one representative chromatographic run per treatment condition; these are acknowledged limitations of the present dataset (Section 3.8) rather than a statistical claim of replicate variance. GC-MS peak-area percentage was treated throughout as a semi-quantitative measure of relative abundance within a chromatographic run, and compound identification

was accepted only where the observed retention time and mass spectrum matched the reference library entry and CAS registry number; peaks falling below the library identification threshold were reported as undetectable rather than assigned a nominal zero concentration.

3. Results and Discussion

3.1 Surface Morphology (SEM)

Scanning electron micrographs of unmodified periwinkle-derived chitosan and the chitosan–lemon waste graft (CLWG) are presented in Fig. 1a and Fig. 1b, respectively (300× magnification, 883 µm field of view, 15 kV, backscattered-detector full mode).

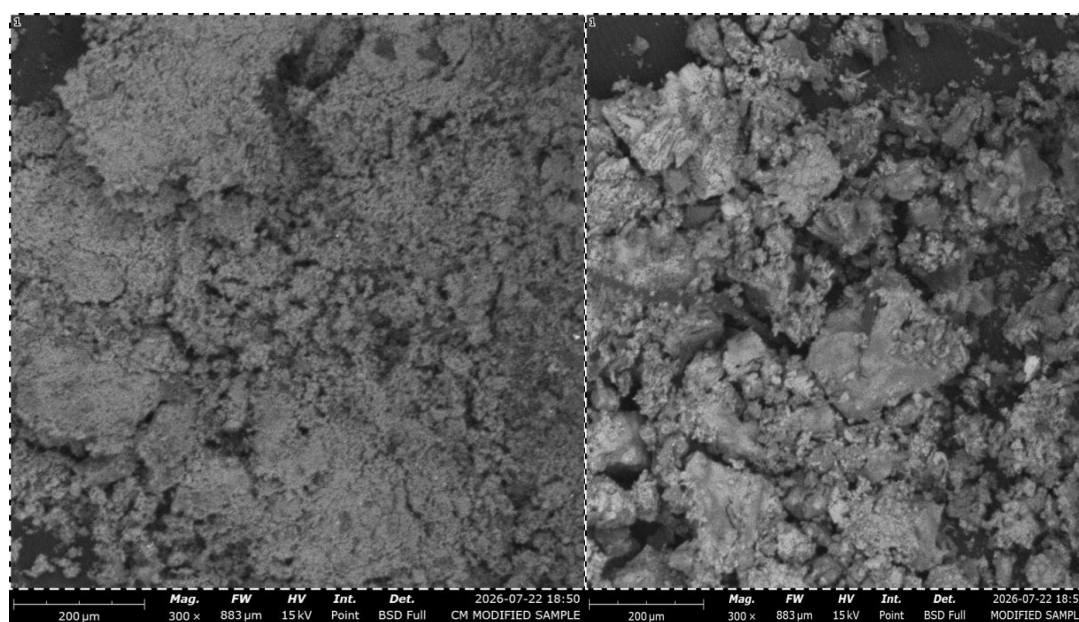


Fig. 1 Left: Scanning electron micrograph of unmodified periwinkle-derived chitosan (300×, 883 µm FOV, 15 kV)
Right: Scanning electron micrograph of the chitosan–lemon waste graft (CLWG) (300×, 883 µm FOV, 15 kV)

The unmodified periwinkle-derived chitosan (Fig. 1a) displays a compact, fine-grained, and relatively homogeneous surface with densely packed aggregates and only limited visible porosity, consistent with the mineral-reinforced surface architecture reported for periwinkle- and snail-shell-derived chitosan, in which residual calcium carbonate from incomplete demineralisation imparts a smooth, tightly packed particle texture [8,10]. After grafting, the CLWG (Fig. 1b) shows a markedly different morphology: larger, angular, blocky particle domains with sharper edges and visible fracture planes, interspersed with irregular voids and a generally coarser, more heterogeneous texture than the unmodified precursor. This shift from a smooth, fine aggregate morphology to a rougher, more structurally heterogeneous surface is the classical SEM signature of successful chitosan grafting or compositing reported elsewhere: Abd El-Sayed *et al.* [35] observed that SEM revealed “intensive covering” of chitosan onto a dialdehyde cellulose backbone after grafting, and similar coarsening and roughening on

grafting has been reported for chitosan-poly(ortho-toluidine) and chitosan-poly(N-vinylpyrrolidone) graft copolymers [36]. Critically, the CLWG morphology does not resemble either the unreacted chitosan precursor or two cleanly segregated phases that would be expected of a simple physical blend of chitosan and carbonised lemon waste; instead, the integrated, interpenetrating rough matrix observed is more consistent with a genuine composite in which the lemon-waste-derived carbon/citrate phase is intimately associated with the chitosan backbone. This interpretation is directly corroborated by the elemental and spectroscopic evidence discussed in Sections 3.2 and 3.3. The increased surface heterogeneity and apparent porosity of the CLWG relative to unmodified chitosan is also functionally significant: rougher, more textured adsorbent surfaces generally expose a greater density of accessible binding sites, a structure-function relationship widely invoked to explain the enhanced pollutant uptake of grafted or composited chitosan adsorbents relative to the native polymer [19,20,37].

3.2 Elemental Composition (EDX)

Table 1 and Table 2 present the EDX-derived elemental composition (atomic % and weight %) of unmodified chitosan and the CLWG, respectively, with the corresponding spectra shown in Fig. 2a and Fig. 2b.

Table 1 EDX elemental composition of unmodified chitosan

Element No.	Symbol	Element Name	Atomic Conc. (%)	Weight Conc. (%)
20	Ca	Calcium	78.22	77.54
30	Zn	Zinc	6.09	9.84
17	Cl	Chlorine	5.28	4.63
14	Si	Silicon	4.77	3.32
25	Mn	Manganese	2.01	2.73
15	P	Phosphorus	1.82	1.40
6	C	Carbon	1.81	0.54
12	Mg	Magnesium	0.00	0.00
19	K	Potassium	0.00	0.00
22	Ti	Titanium	0.00	0.00
26	Fe	Iron	0.00	0.00
13	Al	Aluminium	0.00	0.00
16	S	Sulfur	0.00	0.00
7	N	Nitrogen	0.00	0.00

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Table 2 EDX elemental composition of the chitosan–lemon waste graft (CLWG)

Element No.	Symbol	Element Name	Atomic Conc. (%)	Weight Conc. (%)
17	Cl	Chlorine	54.67	55.35
26	Fe	Iron	8.97	14.31
14	Si	Silicon	16.56	13.28
13	Al	Aluminium	4.47	3.45
30	Zn	Zinc	1.70	3.17
25	Mn	Manganese	1.83	2.87
20	Ca	Calcium	2.21	2.53
22	Ti	Titanium	1.46	2.00
7	N	Nitrogen	4.65	1.86
6	C	Carbon	3.47	1.19
12	Mg	Magnesium	0.00	0.00
19	K	Potassium	0.00	0.00
15	P	Phosphorus	0.00	0.00
16	S	Sulfur	0.00	0.00

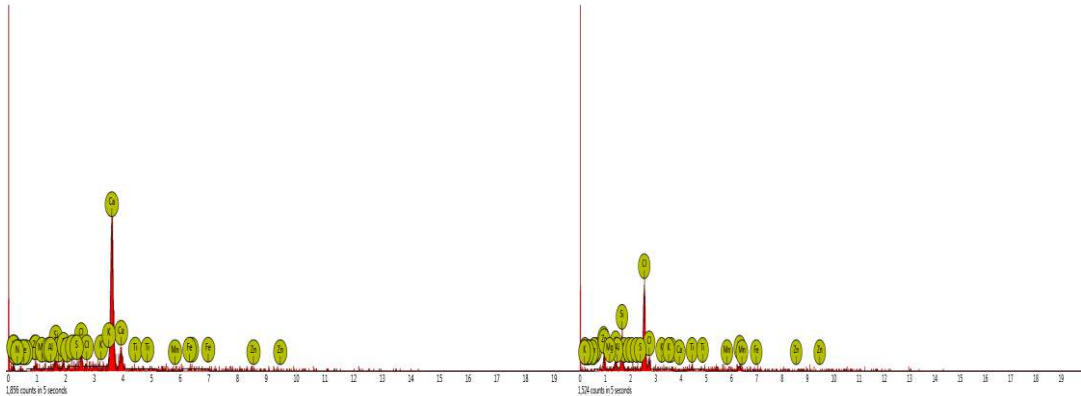


Fig. 2 Left: EDX spectrum of unmodified chitosan (FOV: 883 μm , 15 kV, Point mode, BSD Full)

Fig. 2 Right: EDX spectrum of the chitosan–lemon waste graft (CLWG) (FOV: 883 μm , 15 kV, Point mode, BSD Full)

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286
287

288 The EDX spectra provide the most direct elemental confirmation of grafting. Unmodified chitosan
289 (Table 1, Fig. 2a) is dominated by calcium (78.22 atomic %), with smaller contributions from zinc,
290 chlorine, silicon, manganese, phosphorus, and carbon, and, notably, no detectable nitrogen (0.00
291 atomic %). This calcium dominance reflects the native mineral architecture of the periwinkle shell
292 precursor: Gbenebor *et al.* [8] reported a rough, mineral-reinforced surface for periwinkle- and snail-
293 derived chitin/chitosan attributable to residual shell calcium carbonate, and Ewii *et al.* [10] similarly
294 reported elevated ash content (10.5%) in periwinkle-derived chitosan relative to crab- and snail-derived
295 material, reflecting incomplete removal of the mineral phase during demineralisation. The apparent
296 absence of nitrogen in the unmodified sample should not be read as an absence of the chitosan
297 biopolymer itself; nitrogen K α X-rays are low in energy and disproportionately prone to self-absorption
298 and reduced detection efficiency in EDX, particularly under point-mode acquisition sampling a highly
299 mineral-dominated near-surface region [32,33], so the calcium-rich shell-derived surface layer most
300 likely masked the underlying nitrogenous polymer at the specific point analysed. After grafting, the
301 CLWG spectrum (Table 2, Fig. 2b) changes substantially: calcium falls from 78.22 to 2.21 atomic %,
302 while chlorine rises sharply to become the dominant element (54.67 atomic %), and iron (8.97%),
303 silicon (16.56%), aluminium (4.47%), and titanium (1.46%) all appear at levels not detected in the
304 unmodified sample. Most diagnostically, nitrogen — undetectable in the unmodified precursor — is
305 now clearly present at 4.65 atomic %. This emergence of nitrogen after grafting is the same signature
306 reported by Abd El-Sayed *et al.* [35], in which EDX “proved the emergence of nitrogen derived from
307 chitosan” as confirmatory evidence of successful grafting onto a non-nitrogenous backbone; here, the
308 pattern is consistent with the acidic dissolution and citric-acid crosslinking steps of the CLWG

synthesis (2% HCl dissolution of chitosan; citric acid treatment of the lemon waste precursor) dissolving and displacing much of the native calcium carbonate shell mineral, thereby exposing a larger relative proportion of the nitrogenous chitosan backbone and the grafted lemon-waste phase at the analysed surface. The sharp rise in chlorine is plausibly attributable to residual chloride from the HCl-mediated dissolution step and its incomplete removal during the pH-adjustment and washing stages of synthesis, while the appearance of Fe, Al, Si, and Ti most likely reflects trace mineral or clay-associated impurities intrinsic to the lemon waste precursor becoming proportionally more prominent once the dominant calcium signal is reduced. Silicon-bearing residues are also increasingly recognised as ubiquitous environmental contaminants originating from siloxane-based consumer and industrial materials [38], and cannot be entirely excluded as a contributing source, although the co-occurrence of Si with Fe, Al, and Ti here is more consistent with a mineral/clay origin than with siloxane contamination specifically. Because EDX in point-analysis mode interrogates a very small, potentially non-representative surface region, some caution is warranted before over-interpreting the exact magnitude of each elemental shift; area-mapping EDX across multiple particle regions, rather than single-point acquisition, would provide a more representative and statistically robust confirmation of bulk elemental change in future work. Nonetheless, the appearance of nitrogen alongside the collapse of the calcium signal is, taken together with the morphological change documented in Section 3.1, strong converging evidence that grafting was chemically successful rather than a simple unreacted physical mixture.

3.3 Functional Group Analysis (FTIR)

ATR-FTIR spectra (4000–400 cm^{-1}) of unmodified chitosan, the CLWG, and the dried spent CLWG recovered from the highest-dose, longest-contact (1.0 g/8 h) adsorption trial are shown in Fig. 3a–c.

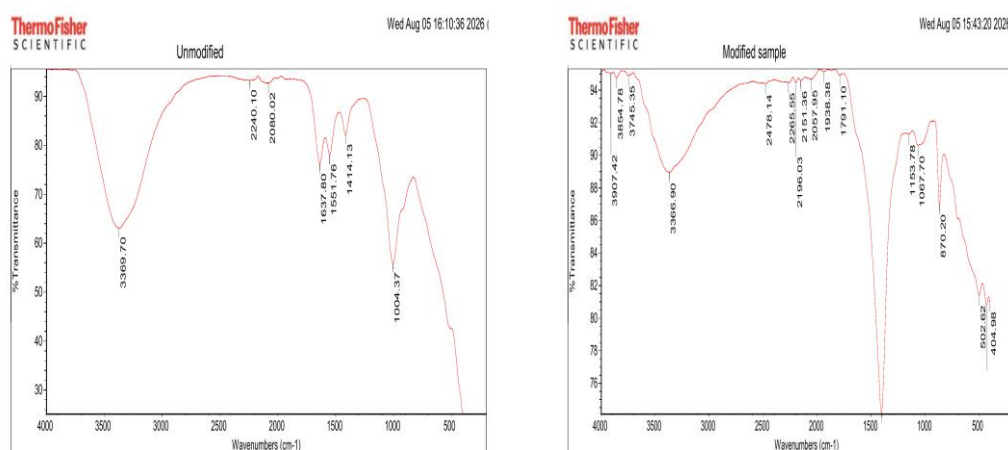


Fig. 3a Left: ATR-FTIR spectrum of unmodified chitosan. **Right:** ATR-FTIR spectrum of the chitosan–lemon waste graft (CLWG)

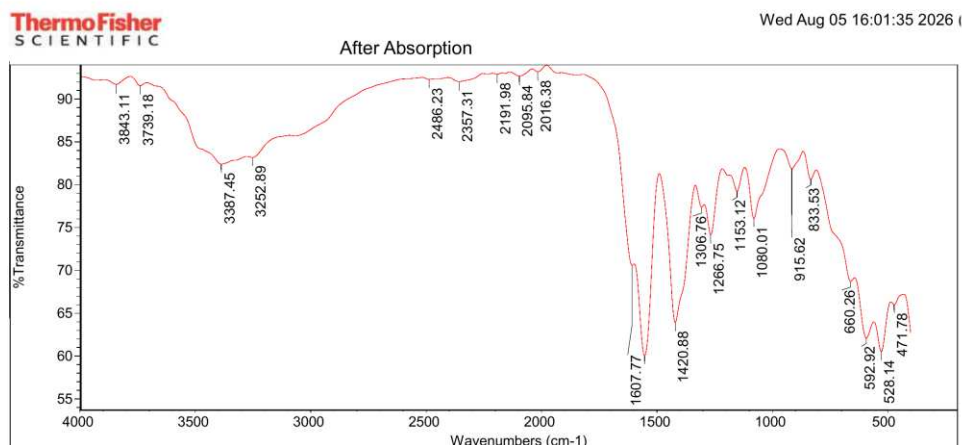


Fig. 3b ATR-FTIR spectrum of the CLWG after adsorption (1.0 g/8 h batch)

The ATR-FTIR spectrum of unmodified chitosan (Fig. 3a) shows the expected fingerprint of the native biopolymer: a broad band at 3369.70 cm^{-1} corresponding to overlapping O–H and N–H stretching vibrations, an Amide I band (C=O stretch of residual N-acetyl groups) at 1637.80 cm^{-1} , an Amide II band (N–H bending) at 1551.76 cm^{-1} , a band at 1414.13 cm^{-1} attributable to CH_2/CH_3 deformation, and a broad skeletal band at 1004.37 cm^{-1} corresponding to the C–O–C glycosidic bridge and pyranose ring vibrations. This pattern closely matches the diagnostic chitosan bands reported by Kasaai [39] and Brugnerotto *et al.* [40], and the O–H/N–H, glycosidic, and amide signatures reported for periwinkle-derived chitosan and chitin by Onosakponome *et al.* [9]. After grafting, the CLWG spectrum (Fig. 3b) retains the O–H/N–H envelope at a very similar position (3366.90 cm^{-1}), but shows a new, strong, and sharply saturated absorption in the $1500\text{--}1600\text{ cm}^{-1}$ region that dominates the spectrum — substantially more intense than the corresponding Amide I/II bands of the unmodified precursor. This new intense carbonyl/carboxylate-region absorption is consistent with the formation of new ester and amide linkages between the citric acid crosslinker and the chitosan amine/hydroxyl groups during synthesis, a mechanism well documented for citric-acid-crosslinked chitosan adsorbents, in which new carbonyl and carboxylate bands appear in this same spectral region upon crosslinking [41]. The CLWG spectrum also shows a cluster of new weak combination bands between 1791 and 2478 cm^{-1} that are absent from the unmodified spectrum, together with altered fine structure in the glycosidic C–O–C region (new resolved bands at 1153.78 and 1067.70 cm^{-1} , compared with the single broad 1004.37 cm^{-1} band of unmodified chitosan) and a new band at 870.20 cm^{-1} plausibly reflecting incorporation of aromatic or anomeric C–H character from the lignocellulosic lemon-waste component. Taken together, the appearance of a new dominant carbonyl-region absorption, the altered glycosidic fine structure, and the new low-frequency bands (502.62 and 404.98 cm^{-1}) constitute spectroscopic evidence of covalent chemical modification rather than a simple physical mixture, corroborating the nitrogen-emergence finding from EDX (Section 3.2) and the morphological change observed by SEM (Section 3.1); the convergence of all three independent lines of evidence on the same conclusion is the standard multi-technique confirmation strategy for chitosan graft/composite formation [12]. The post-adsorption spectrum of the spent 1.0 g/8 h CLWG (Fig. 3c) differs further still. The single broad O–H/N–H envelope of the pre-adsorption CLWG resolves into two distinct, better-defined peaks at 3387.45 and

3252.89 cm^{-1} , a classic signature of new, more ordered hydrogen-bonding environments forming between the adsorbent's $-\text{OH}/-\text{NH}_2$ groups and adsorbate functional groups. A strong new band appears at 1607.77 cm^{-1} , in the region associated with aromatic C=C ring stretching and/or N-H bending of a complexed amine, consistent with binding of the aromatic amine pollutants identified by GC-MS (Section 3.5). Additional new or intensified bands at 1420.88, 1306.76, and 1266.75 cm^{-1} fall within the C-N stretching region expected for amine-adsorbent coordination, while two new bands at 915.62 and 833.53 cm^{-1} , in the aromatic C-H out-of-plane bending region, are consistent with the substitution pattern of an adsorbed substituted benzenamine/aniline-type ring. Finally, a cluster of new low-frequency bands (660.26, 592.92, 528.14, and 471.78 cm^{-1}) falls within the range typically associated with As-O stretching and metal-oxygen coordination vibrations, consistent with surface complexation of the arsenous-acid-derived organoarsenic species also identified in the untreated wastewater (Section 3.5). This overall pattern of band splitting in the O-H/N-H region combined with new bands in the amine-, aromatic-, and metal-oxide-coordination regions indicates that the CLWG's hydroxyl and amine functional groups were directly and simultaneously involved in binding both organoarsenic and aromatic amine pollutants, mechanistically consistent with the hydrogen-bonding, electrostatic, and coordination mechanisms reported for chitosan-based sorbents targeting arsenic [42,43] and aromatic amines [23] individually.

3.4 GC-MS Profiling of Untreated and Treated Wastewater

Total ion chromatograms for the untreated wastewater and the four CLWG-treated filtrates (0.5 g/4 h, 0.5 g/8 h, 1 g/4 h, 1 g/8 h) are shown in Fig. 4a–e. The full set of GC-MS-identified organic compounds for each condition, including peak number (PK), retention time (RT, min), library-matched compound identity, CAS registry number, and relative peak area (%), is provided in Additional File 1.

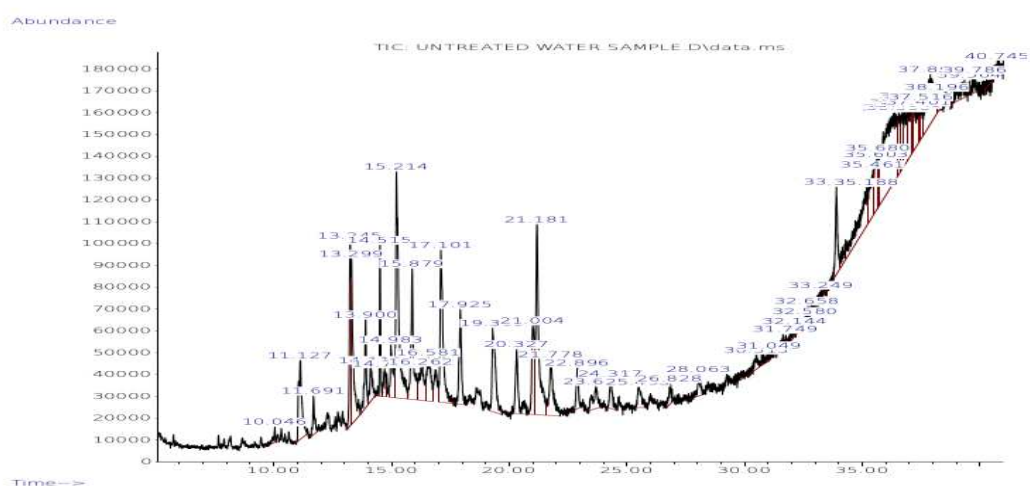
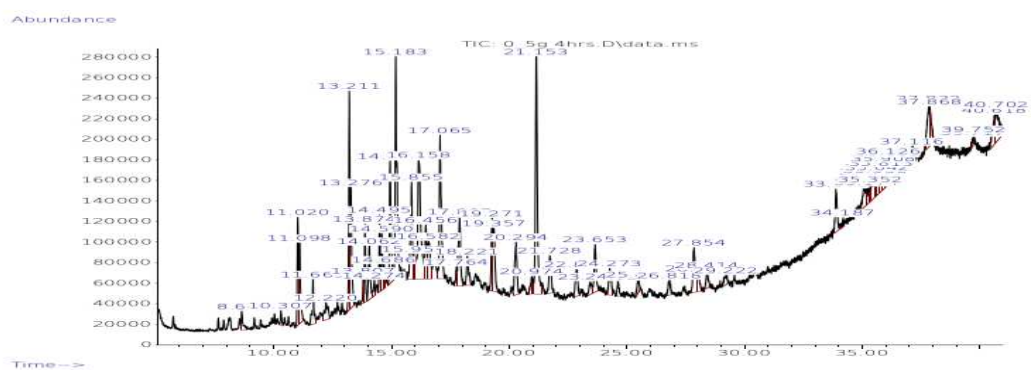


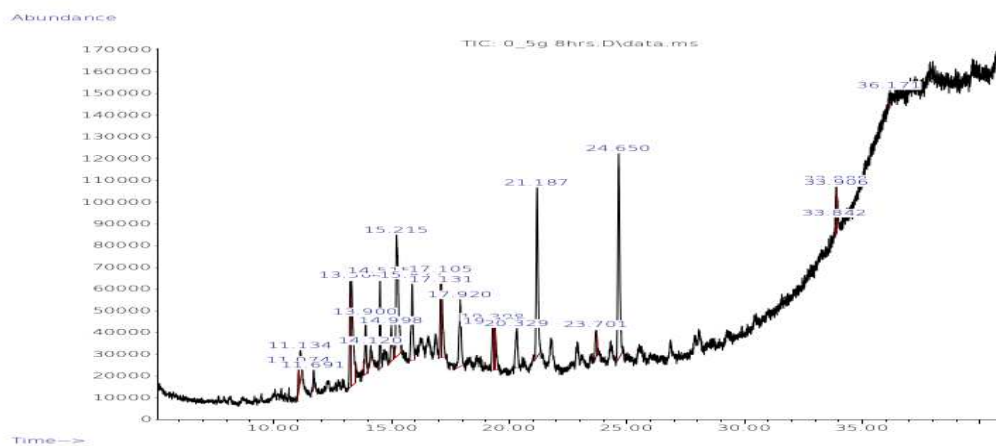
Fig. 4a Total ion chromatogram of untreated wastewater



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Fig. 4b Total ion chromatogram of wastewater treated with 0.5 g CLWG for 4 h



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Fig. 4c Total ion chromatogram of wastewater treated with 0.5 g CLWG for 8 h

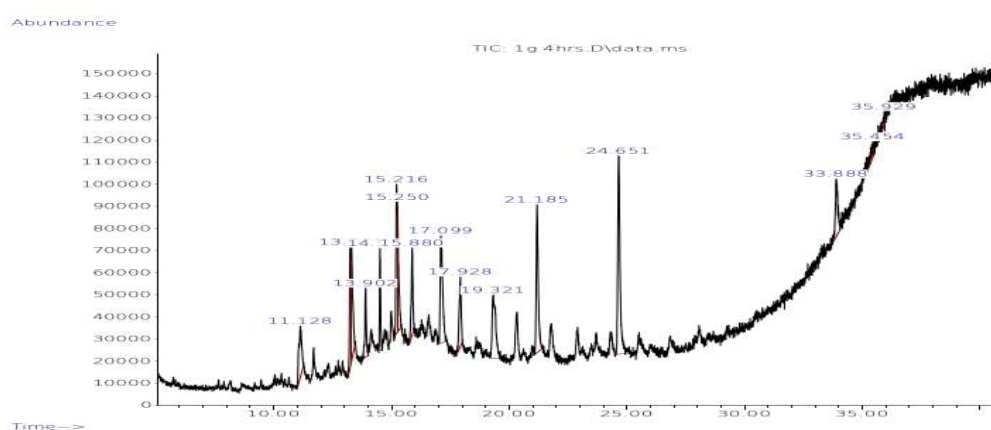


Fig. 4d Total ion chromatogram of wastewater treated with 1 g CLWG for 4 h

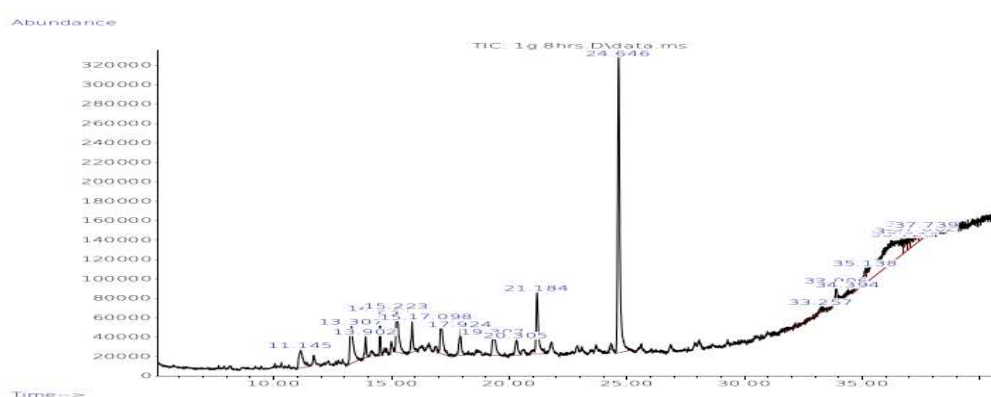


Fig. 4e Total ion chromatogram of wastewater treated with 1 g CLWG for 8 h

3.5 Organoarsenic Species and Aromatic Amine Compounds Identified by GC-MS

To directly evaluate removal of the two pollutant classes targeted in this study, the organoarsenic and aromatic-amine-class compounds identified within each GC-MS run (Additional File 1) were extracted and are presented individually in Table 3 (untreated wastewater) and Table 4 (the one treated condition with detectable species), one per treatment condition, showing peak number, retention time, compound identity, CAS registry number, and relative peak area (%). Conditions with no organoarsenic or aromatic amine species detected above the instrument identification threshold are reported as such rather than with an empty table.

Table 3 Organoarsenic and aromatic amine species identified in untreated wastewater

PK	RT (min)	Compound	CAS No.	Area %	Class
19	21.0043	Benzenamine, 2,6-diethyl-	000579-66-8	3.0392	Aromatic amine
37	35.4615	N,N-Dimethyl-4-nitroso-3-(trimethylsilyl)aniline	017993-84-9	1.8814	Aromatic amine
44	37.0430	Arsenous acid, tris(trimethylsilyl) ester	055429-29-3	1.5168	Organoarsenic

Table 4 Organoarsenic and aromatic amine species identified after treatment with 0.5 g CLWG for 4 hours

PK	RT (min)	Compound	CAS No.	Area %	Class
47	35.1738	4H-1,2,4-triazole-3,5-diamine, N3-(4-fluorophenyl)-N5-methyl-	1000396-10-8	0.5224	Aromatic amine
50	35.5584	2-Ethylacridine	055751-83-2	0.3622	N-heterocyclic aromatic
52	35.8129	2-Ethylacridine	055751-83-2	0.6568	N-heterocyclic aromatic
54	36.0249	2-Ethylacridine	055751-83-2	0.8577	N-heterocyclic aromatic

Organoarsenic and aromatic amine species identified after treatment with 0.5 g CLWG for 8 hours — no organoarsenic or aromatic amine species were identified above the instrument identification threshold.

Organoarsenic and aromatic amine species identified after treatment with 1 g CLWG for 4 hours — no organoarsenic or aromatic amine species were identified above the instrument identification threshold.

Organoarsenic and aromatic amine species identified after treatment with 1 g CLWG for 8 hours — no organoarsenic or aromatic amine species were identified above the instrument identification threshold.

3.6 Removal of the Organoarsenic Species

Only one organoarsenic species was identified by GC-MS across all five chromatographic runs: arsenous acid, tris(trimethylsilyl) ester (CAS 055429-29-3), detected exclusively in the untreated wastewater (Table 3) at a relative peak area of 1.5168% (RT 37.0430 min, peak 44). This compound is the trimethylsilyl-derivatised form of arsenous acid (arsenite), the trivalent inorganic arsenic species described in the Introduction as markedly more toxic and mobile than pentavalent or methylated organic arsenicals [2,3]; its silylated appearance in the chromatogram is an expected artefact of the sample matrix rather than evidence of a genuinely organic arsenic compound, since silylating agents and residual TMS-active species in complex environmental extracts routinely derivatise labile protic analytes, including arsenous acid, into their trimethylsilyl esters prior to or during GC-MS analysis [44]. Its detection in the untreated wastewater therefore functionally confirms the presence of bioavailable arsenite in the raw effluent, consistent with the manuscript's premise that industrial discharge is a significant anthropogenic vector for arsenic entry into receiving waters [1]. Critically, this organoarsenic signal is undetectable in every treated condition (Table 4 and Section 3.5), including the lowest tested dose and shortest tested contact time (0.5 g/4 h). No new arsenic-containing species appeared at any dose or contact time. This indicates that the CLWG achieved apparent complete removal of the detectable organoarsenic burden under all four tested treatment conditions, with no evidence of a dose- or time-dependent removal gradient because the species was already below the GC-

MS identification threshold at the least intensive treatment condition tested. This is consistent with the amine- and hydroxyl-mediated arsenic-binding mechanism documented for other chitosan-based sorbents, in which protonated amine and hydroxyl groups on the chitosan backbone act as ligands for arsenite/arsenate complexation [42,43], and is corroborated here by the new As–O-region FTIR bands observed in the post-adsorption spectrum (Section 3.3). Because only a single organoarsenic species was detected at only one condition, this result should be interpreted as a strong preliminary indication rather than a fully quantified removal efficiency; confirmatory replicate analysis and, ideally, a validated arsenic-specific quantitative method (e.g., ICP-MS or hydride-generation AAS) would be needed to establish a numerical percentage removal with statistical confidence.

3.7 Removal of the Aromatic Amine Compounds

Two aromatic amine species were identified in the untreated wastewater (Table 3). Benzenamine, 2,6-diethyl- (CAS 000579-66-8) was the most abundant, at 3.0392% relative peak area (RT 21.0043 min, peak 19); this is a simple ring-substituted aniline of the type widely used as a dye and rubber-chemical intermediate, and its presence is consistent with the effluent's origin in an industrial area associated with textile and allied azo-dye processing, where aniline-derived intermediates are a recognised source of aromatic amine residues [4,6]. N,N-Dimethyl-4-nitroso-3-(trimethylsilyl)aniline (CAS 017993-84-9) was also identified, at 1.8814% (RT 35.4615 min, peak 37); this is a nitrosated, silylated aniline derivative in which the trimethylsilyl group is again attributable to matrix/derivatisation artefact rather than a genuine structural feature of the parent pollutant [44], but the nitroso substitution on the aniline ring is toxicologically notable, since N-nitroso aromatic amine derivatives are recognised as more potent mutagens than their parent anilines [5]. Both aromatic amine species were undetectable in every treated condition (Table 4 and Section 3.5), indicating apparent complete removal at every tested dose and contact time, consistent with the amine-selective binding capacity previously demonstrated for aniline-functionalised chitosan adsorbents [23] and corroborated by the new aromatic C=C and C–N bands observed in the post-adsorption FTIR spectrum (Section 3.3). A more complex pattern emerged at the lowest dose and shortest contact time (0.5 g/4 h; Table 4), where neither original aromatic amine was detected, but two new nitrogen-containing aromatic species appeared that were absent from the untreated baseline: 4H-1,2,4-triazole-3,5-diamine, N3-(4-fluorophenyl)-N5-methyl- (CAS 1000396-10-8, 0.5224%, RT 35.1738 min) and 2-ethylacridine (CAS 055751-83-2), which recurred at three separate retention times (0.3622%, 0.6568%, and 0.8577%, totalling 1.8767% combined peak area). Because neither compound is present in the untreated wastewater, they cannot represent surviving parent pollutant; two explanations are plausible. First, they may represent partial transformation products of the original aniline-type pollutants, generated during the 4-hour contact period: acridines are classically formed via oxidative cyclisation/self-condensation of aniline-type precursors, a pathway that could plausibly be promoted at the CLWG surface by the transition-metal content (Fe, Mn, Ti) revealed by EDX (Section 3.2). Second, they may represent trace nitrogenous heterocycles intrinsic to the CLWG precursor materials themselves (carbonised lemon waste and periwinkle shell) that desorb into solution under the least intensive treatment condition before being themselves captured at higher doses or longer contact times. It is also worth noting that acridine is a fused-ring tertiary nitrogen

heterocycle rather than a primary or secondary exocyclic aromatic amine in the strict sense, so its classification alongside the aromatic amines targeted by this study is presented here for transparency and completeness rather than as an unambiguous parent-pollutant match. Both transient species were, in any case, undetectable at every higher dose and/or longer contact time (Section 3.5), indicating that increasing either the CLWG dose (to 1 g) or the contact time (to 8 h) was sufficient to eliminate not only the two original aromatic amine pollutants but also these transient low-dose byproducts.

3.8 Overall Removal Pattern and Study Limitations

Across all five GC-MS runs, a consistent pattern emerges: the organoarsenic species and both original aromatic amine compounds identified in the untreated wastewater were undetectable at every tested CLWG dose and contact time, including the least intensive condition (0.5 g/4 h), while the only detectable nitrogen-aromatic byproducts at that lowest-intensity condition were themselves eliminated once either dose or contact time was increased. No organoarsenic or aromatic amine species of any kind were detected at 0.5 g/8 h, 1 g/4 h, or 1 g/8 h (Section 3.5), indicating that apparent complete removal of both target pollutant classes is achievable well within the tested experimental range. This outcome is broadly consistent with reports that chitosan-based sorbents achieve high removal efficiencies for both arsenic oxyanions [42,43] and aromatic amine/aniline-type pollutants [23] individually, and extends this evidence to a dual-agro-waste-valorised graft evaluated against real, uncharacterised industrial effluent rather than a synthetic single-pollutant solution. Several limitations should nonetheless be acknowledged. GC-MS peak-area percentage is a semi-quantitative measure of relative abundance within a chromatographic run rather than a validated, calibrated concentration; a compound falling below the library identification threshold indicates removal below instrumental detectability under the conditions used here, not necessarily removal to zero absolute concentration, and confirmatory quantitative analysis (e.g., matrix-matched calibration curves, or orthogonal techniques such as ICP-MS for the arsenic species) would strengthen this conclusion. The batch adsorption trials were conducted in duplicate, but the GC-MS profiling reported here reflects one representative chromatographic run per condition; replicate GC-MS analysis would allow the transient low-dose byproducts identified in Section 3.7 to be distinguished with greater confidence from genuine single-run artefacts. Finally, because EDX was conducted in point-analysis mode at a single surface location per sample (Section 3.2), the elemental composition data, while diagnostic, should be corroborated in future work by area-mapping across multiple particle regions to confirm that the reported elemental shifts are representative of the bulk material rather than local surface heterogeneity.

4. Conclusions

This study valorised two co-located agro-industrial waste streams — periwinkle shell and lemon fruit waste — into a single biofunctionalized adsorbent, the chitosan–lemon waste graft (CLWG), and evaluated its performance against real, uncharacterised industrial wastewater rather than a synthetic single-pollutant solution. Three independent lines of evidence converged to confirm successful grafting: SEM showed a shift from the compact, fine-grained morphology of unmodified chitosan to a coarser, more heterogeneous surface after modification; EDX confirmed the emergence of nitrogen

(0.00 to 4.65 atomic %) alongside a sharp decline in calcium (78.22 to 2.21 atomic %), consistent with exposure of the chitosan-derived nitrogenous phase following acid-mediated dissolution of the native shell mineral; and ATR-FTIR revealed a new dominant carbonyl-region absorption and altered glycosidic fine structure consistent with citric-acid-mediated crosslinking. GC-MS profiling of the raw effluent identified one organoarsenic species (arsenous acid, tris(trimethylsilyl) ester) and two aromatic amine compounds (benzenamine, 2,6-diethyl-, and N,N-dimethyl-4-nitroso-3-(trimethylsilyl)aniline); all three were undetectable at every tested CLWG dose and contact time (0.5–1.0 g; 4–8 h), and two transient nitrogen-aromatic byproducts observed only at the lowest dose and shortest contact time were themselves absent once either dose or contact time was increased. Post-adsorption FTIR band splitting and the appearance of new amine-, aromatic-, and metal-oxide-coordination bands provided direct spectroscopic evidence that the CLWG's hydroxyl and amine functional groups were mechanistically involved in binding both pollutant classes. Collectively, these findings indicate that a low-cost, dual-agro-waste-valorised chitosan graft can achieve apparent complete removal of both organoarsenic species and aromatic amine compounds from real industrial wastewater, supporting its potential as a circular-economy remediation strategy for resource-limited settings where periwinkle shell and citrus waste are regionally abundant. This aligns with a broader shift toward resource-recovery-oriented wastewater management, exemplified by recent efforts to reclaim high-value bioproducts such as squalene from oilfield wastewater streams [45], in which waste valorisation and pollutant remediation are pursued jointly rather than as separate objectives. Future work should prioritise replicate and quantitatively calibrated GC-MS or ICP-MS confirmation of removal efficiency, area-mapping EDX to corroborate the point-analysis elemental findings reported here, and isotherm/kinetic modelling of the adsorption process, for example using the pseudo-second-order framework of Ho and McKay [46], across a wider dose and contact-time range.

Declarations

Ethics approval and consent to participate

Not applicable. This study did not involve human participants, human data, or animals; it involved laboratory-based characterisation of a biosorbent material and analysis of industrial wastewater samples only.

Consent for publication

Not applicable.

Availability of data and materials

The GC-MS raw instrument files, SEM images, EDX spectra, and ATR-FTIR spectra generated and analysed during the current study, together with the treatment-condition crosswalk (untreated, 0.5 g/4 h, 0.5 g/8 h, 1 g/4 h, 1 g/8 h; Tables 1–4 and Additional File 1), are available from the corresponding author upon reasonable request.

Competing interests

The authors declare no competing financial or non-financial interests.

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Authors' contributions

Mathew Gideon: Conceptualization, methodology, investigation, formal analysis, writing – original draft, writing – review and editing. The author read and approved the final manuscript.

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List of Abbreviations

CLWG, chitosan–lemon waste graft; SEM, scanning electron microscopy; EDX, energy-dispersive X-ray spectroscopy; ATR-FTIR, attenuated total reflectance Fourier-transform infrared spectroscopy; GC-MS, gas chromatography–mass spectrometry; WHO, World Health Organization; IARC, International Agency for Research on Cancer; CAS, Chemical Abstracts Service (registry number); RT, retention time; PK, peak number; HCl, hydrochloric acid; NaOH, sodium hydroxide.

References

1. Karagas MR, Gossai A, Pierce B, Ahsan H. Drinking water arsenic contamination, skin lesions, and malignancies: A systematic review of the global evidence. *Current Environmental Health Reports*. 2015;2(1):52–68. Available from: <https://doi.org/10.1007/s40572-014-0040-x>
2. Eisler R. Arsenic hazards to fish, wildlife, and invertebrates: A synoptic review (Biological Report 85(1.12), Contaminant Hazard Reviews Report No. 12). U.S. Fish and Wildlife Service. 1988.
3. World Health Organization. Arsenic [Internet]. 2022. Available from: <https://www.who.int/news-room/fact-sheets/detail/arsenic>
4. Vineis P, Pirastu R. Aromatic amines and cancer. *Cancer Causes & Control*. 1997;8(3):346–355. Available from: <https://doi.org/10.1023/A:1018453104303>
5. Arora PK. Bacterial degradation of monocyclic aromatic amines. *Frontiers in Microbiology*. 2015;6:Article 820. Available from: <https://doi.org/10.3389/fmicb.2015.00820>

- 579 6. Edebali Ö, Krupčíková S, Goellner A, Vrana B, Muz M, Melymuk L. Tracking aromatic
580 amines from sources to surface waters. *Environmental Science & Technology Letters*.
581 2024;11(5):397–409. Available from: <https://doi.org/10.1021/acs.estlett.4c00032>
- 582 7. Richardson SD, Ternes TA. Water analysis: Emerging contaminants and current issues.
583 *Analytical Chemistry*. 2014;86(6):2813–2848. Available from:
584 <https://doi.org/10.1021/ac500508t>
- 585 8. Gbenebor OP, Akpan EI, Adeosun SO. Thermal, structural and acetylation behavior of snail
586 and periwinkle shells chitin. *Progress in Biomaterials*. 2017;6(3):97–104. Available from:
587 <https://doi.org/10.1007/s40204-017-0070-1>
- 588 9. Onosakponome I, Okwuenu PC, Oparaji EH, Isojie AO, Odesa GE, Awhin EP, et
589 al. Extraction and FTIR spectra of chitin and chitosan produced from periwinkle
590 (*Tympanotonos fuscatus*) under differential decolorization conditions. *Tropical Journal of*
591 *Chemistry*. 2025;1(1):50–57. Available from: <https://doi.org/10.71148/tjoc/v1i1.7>
- 592 10. Ewii UE, Olorunsola EO, Onugwu AL, Ameh OM, Eke EN, Iheme CI, et al. A comparative
593 study on the physicochemical characterization of chitosan obtained from seafood in Uyo
594 metropolis, Nigeria. *Tropical Journal of Natural Product Research*, 9(7). 2025. Available
595 from: <https://doi.org/10.26538/tjnpr/v9i7.60>
- 596 11. Varma R, Vasudevan S. Extraction, characterization, and antimicrobial activity of chitosan
597 from horse mussel *Modiolus modiolus*. *ACS Omega*. 2020;5(32):20224–20230. Available
598 from: <https://doi.org/10.1021/acsomega.0c01903>
- 599 12. Andrew EC, Grace EA, Chinemerem OE, Emeka OE. Mini review on the morphological and
600 microanalysis characterization of chitosan nanocomposites using scanning electron
601 microscopy. *Mathews Journal of Pharmaceutical Science*. 2025;9(3):52. Available from:
602 <https://doi.org/10.30654/MJPS.10052>
- 603 13. Bhatnagar A, Sillanpää M. Applications of chitin- and chitosan-derivatives for the
604 detoxification of water and wastewater—A short review. *Advances in Colloid and Interface*
605 *Science*. 2009;152(1–2):26–38. Available from: <https://doi.org/10.1016/j.cis.2009.09.003>
- 606 14. Crini G. Non-conventional low-cost adsorbents for dye removal: A review. *Bioresource*
607 *Technology*. 2006;97(9):1061–1085. Available from:
608 <https://doi.org/10.1016/j.biortech.2005.05.001>
- 609 15. Kyzas GZ, Bikiaris DN. Recent modifications of chitosan for adsorption applications: A
610 critical and systematic review. *Marine Drugs*. 2015;13(1):312–337. Available from:
611 <https://doi.org/10.3390/md13010312>

- 612 16. Saheed IO, Oh WD, Suah FBM. Chitosan modifications for adsorption of pollutants—A
613 review. *Journal of Hazardous Materials*. 2021;408:Article 124889. Available from:
614 <https://doi.org/10.1016/j.jhazmat.2020.124889>
- 615 17. Atangana E, Ajiboye TO, Mafolasire AA, Ghosh S, Hakeem B. Adsorption of organic
616 pollutants from wastewater using chitosan-based adsorbents. *Polymers*. 2025;17(4):Article
617 502. Available from: <https://doi.org/10.3390/polym17040502>
- 618 18. Wan Ngah WS, Teong LC, Hanafiah MAKM. Adsorption of dyes and heavy metal ions by
619 chitosan composites: A review. *Carbohydrate Polymers*. 2011;83(4):1446–1456. Available
620 from: <https://doi.org/10.1016/j.carbpol.2010.11.004>
- 621 19. Fan L, Luo C, Li X, Lu F, Qiu H, Sun M. Fabrication of novel magnetic chitosan grafted with
622 graphene oxide to enhance adsorption properties for methyl blue. *Journal of Hazardous*
623 *Materials*. 2012;215–216:272–279. Available from:
624 <https://doi.org/10.1016/j.jhazmat.2012.02.068>
- 625 20. Okoya AA, Akinyele AB, Amuda OS, Ofioezie IE. Chitosan-coated carbon for removal of
626 chromium (VI) and lead (II) from aqueous solutions. *Annual Research & Review in Biology*.
627 2016;9(1). Available from: <https://doi.org/10.9734/ACSJ/2016/21813>
- 628 21. Salim A, Liu Y, Kennedy JF. Comparative adsorption of phthalate esters on chitosan beads.
629 *Carbohydrate Polymers*. 2010;81(3):640–644. Available from:
630 <https://doi.org/10.1016/j.carbpol.2010.03.024>
- 631 22. Net S, Sempéré R, Delmont A, Paluselli A, Ouddane B. Occurrence, fate, behavior and
632 ecotoxicological state of phthalates in different environmental matrices. *Environmental*
633 *Science & Technology*. 2015;49(7):4019–4035. Available from:
634 <https://doi.org/10.1021/es505233b>
- 635 23. Vafakish B, Wilson LD. Aniline-grafted chitosan for metal ion adsorption and sensing.
636 *Molecules*. 2020;25(5):Article 1052. Available from:
637 <https://doi.org/10.3390/molecules25051052>
- 638 24. Tzereme A, Christodoulou E, Kyzas GZ, Kostoglou M, Bikiaris DN, Lambropoulou DA.
639 Chitosan grafted adsorbents for diclofenac pharmaceutical compound removal from single-
640 component aqueous solutions and mixtures. *Polymers*. 2019;11(3):Article 497. Available
641 from: <https://doi.org/10.3390/polym11030497>
- 642 25. Gubitosa J, Rizzi V, Fini P, Nuzzo S, Cosma P. Chitosan/TiO₂ composite films for the
643 removal of 2,4-dinitrophenol from water. *International Journal of Molecular Sciences*.
644 2023;24(10):Article 8552. Available from: <https://doi.org/10.3390/ijms24108552>

26. Dehghani MH, Karri RR, Alimohammadi M, Nazmara S, Zarei A, Saeedi R. Removal of bisphenol A (BPA) from aqueous solutions using chitosan immobilized nanoscale zero-valent iron. *Journal of Molecular Liquids*. 2020;310:Article 113317. Available from: <https://doi.org/10.1016/j.molliq.2020.113317>
27. Monir TSB, Elsayed NH, Azzam AM, Madkour TM. Novel grafted bio-based chitosan adsorbents for selective pollutant removal. *Polymers*. 2024;16(12):Article 1718. Available from: <https://doi.org/10.3390/polym16121718>
28. Yusuff RO, Sonibare JA. Characterization of textile industries' effluents in Kaduna, Nigeria and pollution implications. *Global NEST Journal*. 2004;6(3):212–221. Available from: <https://doi.org/10.30955/gnj.000284>
29. Osin OA, Yu T, Lin S. Oil refinery wastewater treatment in the Niger Delta, Nigeria: Current practices, challenges, and recommendations. *Environmental Science and Pollution Research*. 2017;24(28):22730–22740. Available from: <https://doi.org/10.1007/s11356-017-0009-z>
30. Kayode OF, Luethi C, Rene ER. Management recommendations for improving decentralized wastewater treatment by the food and beverage industries in Nigeria. *Environments*. 2018;5(3):Article 41. Available from: <https://doi.org/10.3390/environments5030041>
31. Nnaji JC, Amaku JF, Amadi OK, Nwadinobi SI. Evaluation and remediation protocol of selected organochlorine pesticides and heavy metals in industrial wastewater using nanoparticles (NPs) in Nigeria. *Scientific Reports*. 2023;13:Article 2181. Available from: <https://doi.org/10.1038/s41598-023-28761-3>
32. Goldstein JI, Newbury DE, Echlin P, Joy DC, Lyman CE, Lifshin E, et al. *Scanning electron microscopy and X-ray microanalysis* (3rd ed.). Springer. 2003. Available from: <https://doi.org/10.1007/978-1-4615-0215-9>
33. Egerton RF. *Electron energy-loss spectroscopy in the electron microscope* (3rd ed.). Springer. 2011. Available from: <https://doi.org/10.1007/978-1-4419-9583-4>
34. Yang Q, Ji H, Xu Z, Li Y, Wang P, Sun J, et al. Ultra-fast and accurate electron ionization mass spectrum matching for compound identification with million-scale in-silico library. *Nature Communications*. 2023;14:Article 3722. Available from: <https://doi.org/10.1038/s41467-023-39279-7>
35. Abd El-Sayed ES, Dacrory S, Essawy HA, Ibrahim HS, Ammar NS, Kamel S. Sustainable grafted chitosan-dialdehyde cellulose with high adsorption capacity of heavy metal. *BMC Chemistry*. 2023;17:Article 120. Available from: <https://doi.org/10.1186/s13065-023-01035-9>
36. Lavlinskaya MS, Sorokin AV, Mikhaylova AA, Kuznetsov EI, Baidamshina DR, Saranov IA, et al. The low-waste grafting copolymerization modification of chitosan is a promising

679 approach to obtaining materials for food applications. *Polymers*. 2024;16(11):Article 1596.
680 Available from: <https://doi.org/10.3390/polym16111596>

681 37. Vakili M, Rafatullah M, Salamatinia B, Abdullah AZ, Ibrahim MH, Tan KB, et
682 al. Application of chitosan and its derivatives as adsorbents for dye removal from water and
683 wastewater: A review. *Carbohydrate Polymers*. 2014;113:115–130. Available from:
684 <https://doi.org/10.1016/j.carbpol.2014.07.007>

685 38. Wang DG, Norwood W, Alaei M, Byer JD, Brimble S. Review of recent advances in research
686 on the toxicity, detection, occurrence and fate of cyclic volatile methyl siloxanes in the
687 environment. *Chemosphere*. 2013;93(5):711–725. Available from:
688 <https://doi.org/10.1016/j.chemosphere.2012.10.041>

689 39. Kasaai MR. A review of several reported procedures to determine the degree of N-acetylation
690 for chitin and chitosan using infrared spectroscopy. *Carbohydrate Polymers*. 2008;71(4):497–
691 508. Available from: <https://doi.org/10.1016/j.carbpol.2007.07.009>

692 40. Brugnerotto J, Lizardi J, Goycoolea FM, Argüelles-Monal W, Desbrières J, Rinaudo M. An
693 infrared investigation in relation with chitin and chitosan characterization. *Polymer*.
694 2001;42(8):3569–3580. Available from: [https://doi.org/10.1016/S0032-3861\(00\)00713-8](https://doi.org/10.1016/S0032-3861(00)00713-8)

695 41. Ayach J, Duma L, Badran A, Hijazi A, Martinez A, Bechelany M, et al. Enhancing
696 wastewater depollution: Sustainable biosorption using chemically modified chitosan
697 derivatives for efficient removal of heavy metals and dyes. *Materials*. 2024;17(11):Article
698 2724. Available from: <https://doi.org/10.3390/ma17112724>

699 42. Boddu VM, Abburi K, Talbott JL, Smith ED, Haasch R. Removal of arsenic (III) and arsenic
700 (V) from aqueous medium using chitosan-coated biosorbent. *Water Research*.
701 2008;42(3):633–642. Available from: <https://doi.org/10.1016/j.watres.2007.08.014>

702 43. Gerente C, Lee VKC, Le Cloirec P, McKay G. Application of chitosan for the removal of
703 metals from wastewaters by adsorption—Mechanisms and models review. *Critical Reviews in*
704 *Environmental Science and Technology*. 2007;37(1):41–127. Available from:
705 <https://doi.org/10.1080/10643380600729089>

706 44. Zarate E, Boyle V, Rupprecht U, Green S, Villas-Boas SG, Baker P, et al. Fully automated
707 trimethylsilyl (TMS) derivatisation protocol for metabolite profiling by GC-MS. *Metabolites*.
708 2016;7(1):Article 1. Available from: <https://doi.org/10.3390/metabo7010001>

709 45. Suleimen YM, Trofimov AN, et al. Squalene recovery from Uzen oil field wastewater: A
710 novel abiotic resource. *Frontiers in Environmental Chemistry*. 2025;6:Article 1672872.
711 Available from: <https://doi.org/10.3389/fenvc.2025.1672872>

- 712 46. Ho YS, McKay G. Pseudo-second order model for sorption processes. *Process Biochemistry*.
713 1999;34(5):451–465. Available from: [https://doi.org/10.1016/S0032-9592\(98\)00112-5](https://doi.org/10.1016/S0032-9592(98)00112-5)