



RESEARCH ARTICLE

Chimera Patterns of Escherichia coli lineage with Multi Stress Hardening Resistance to Antibiotics and Ultraviolet Irradiation

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ARTICLE INFO	ABSTRACT
Received: May 25, 2024 Accepted: Jun 27, 2024	<p>Frequency and distribution patterns of diverse and versatile E. coli lineage with dynamic pluripotent virulency to different potent microbicidal stressors like pasteurization regimes cascaded by well-equipped genetic plasticity to evolved to a chimeras with forbidden prohibited resistance to different antibiotics and tolerance to ultraviolet irradiation cold pasteurization were resident within tap waters chain especially within purification stations covered and protected by electromagnetic clouds barriers of biofilm in Baghdad as an emergent bioterrorism hazard catastrophically threatening lifestyles in this mess. These emergent entities unveiled in this torment could be terminated by lytic bacteriophages later as a new combating pathway for hurtling decontamination cascaded by sterilization efficacy dynamic module. Augmented conservative biofilm construction modified protocols were dependent from verified gold standard tissue culture plates to phenotypic plasmids twistors cascaded via verified Congo red assay. Antibigram dependent antibiotics susceptibility patterns to different drugs of choices with different generations were assigned by Kirby-Bauer disc diffusion method cascaded by VITEK® 2 SYSTEM displayed MIC and MBC thresholds. An experimental design model was verified for susceptibility patterns to cold pasteurization regime via ultraviolet irradiation decontamination scheme. Problematic forbidden recovery dogmas of E. coli lineage from tap waters chains resident within Al-Karkh and Al-Rusafa sectors cascaded by Human UTI clinical cases in Baghdad. Documentary records data unveiled multi stress patterns of recovered isolates to versatile antibiotics with shifting diversity to tolerate irradiation pasteurization. In conclusions: Contamination of tap waters with multi stress hardening E. coli lineage cascaded with similarity genetic indices and phylogenetic tree ancestral relationships with clinical isolates from Human UTI cases as a forbidden complex sequel.</p>
<p>Keywords</p> E. coli lineage Stress adaptation Antibiotics resistance Ultraviolet irradiation Tap waters UTI	
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INTRODUCTION

Primordial quality special topics concerned with Iraqi waters; salinity decipher attentiveness of waters pollutants instigated cross cascaded agricultural, municipal and industrial events announce return streams into sources of freshwater. Broadminded upsurge deposition of waters salinity occasioned from evolution of agronomy surrounding Tigris and Euphrates. Expanding loads of versatile contaminants sequels evolution of diverse economic population societies. Low quality is aggravated by scarcities of waters levels sequels mega agronomy desertification (SWLRI, 2015; Al-Atta, 2019; <https://water.fanack.com/iraq/water-quality-in-iraq/>, 2023).

Unproper and uncontrolled discharging of wastewater or sewage that mixed with tap waters across teared waters pipelines networks predisposing and causing dangerous biohazard contamination struggling especially with genetically modified microorganisms' resident and deposited within a recalcitrant biofilm. *E. coli* is one of the catastrophic biohazards leading indicator of foodborne and waterborne chainsaw puzzles in which, their contamination or pollution indicate presence of other serious forbidden entities and their toxins in versatile and diverse ecosystems. Microbial quality of aquatic environment in terms of pollution vs contamination is a priority determinant special topic issue for hygienic sanitary of produced domestic tap waters. Fecal coliforms in association with an ESKAPE chain top pathogens especially genetically modified Chimeras represent an emergent bioterrorism struggling. Enteropathogenic (EPEC) vs Shiga toxin Enterohaemorrhagic (EHEC) serotypes lineage considered a predominant invader of man, animals and other timeline interconnected ecosystems like food and waters chains. Sophisticated safeguarding of tap waters becomes an international programed issue (Al-Shammary, 2009; Kanaan & Al-Shammary, 2013; Al-Dulaimi & Younes, 2017; Jam et al., 2018; Mouhamad *et al.*, 2017; Daneshmand *et al.*, 2018; Al-Shammary, 2019; A-INaseri & Al-Shammary, 2020; Favere *et al.*, 2021; Some *et al.*, 2021; Ghafil *et al.*, 2022; Oleiwi & Al-Dabbas, 2022).

Worldwide assessment of microbial quality versus quantity of drinking waters cascaded by verified testing profile of purification plans with infected and carrier individual-workers especially with clinical phases of enteric foodborne illnesses (food poisoning) associated with UTI cases can determine the contamination source and outcomes of waters in terms of frequency and distribution patterns or cross-sectional scanning surveys of targeted forbidden pathogenic denominator and their ancestral phylogeny. Critical evaluation cascaded by contamination susceptibility patterns via zoonotic vs transmissible foodborne and waterborne pathogens is of a priority issue especially during insufficient hygienic processing cascaded by increase wastewater debris and this, in addition to the low levels of Tigris and Euphrates rivers with scarcity of rain, exacerbated the vital lifestyle modern problems of mixed microbial and toxic residues pollution (Al-Shammary, 2009; Kanaan & Al-Shammary, 2013; Al-Dulaimi & Younes, 2017; Mouhamad *et al.*, 2017; Daneshmand *et al.*, 2018; Al-Shammary, 2019; Al-Naseri & Al-Shammary, 2020; Favere *et al.*, 2021; Rashid et al., 2023; Some *et al.*, 2021; Ghafil *et al.*, 2022; Oleiwi & Al-Dabbas, 2022). Underprivileged microbial water eminence endangers the lifestyle and care of human, animals as well as diet twisted by aquaculture farmsteads.

Conservative and semiconservative microbial defense mechanisms represent a battle bridge arms race between bacterial immune ecosystem and ecofriendly lytic bacteriophages, deciphered as diverse cyclic oligonucleotide based antiphage signaling system (CBASS) and pyrimidine cyclase system for antiphage resistance (Pycsar) brain cell-suicide machine in diverse bacteria that use cyclic nucleotide signals to induce cell death and prevent viral propagation. Diverse intelligent "CBASS & Pycsar" cascaded components in association with Clustered Regularly Interspaced Short Palindromic Repeats or "CRISPR-CAS and Thoeris" ecosystems represent the main antiphage defense strategies to overcome or combat viral invasion. CBASS orchestrated in cascaded dependent manner to activate in different pathways a programmed cell death or induced apoptosis to prevent phage propagation throughout a cascaded series of DNA degradation, membrane destruction and nicotinamide dinucleotide (NAD) depletion thus preventing spread of viruses' machinery molecules. While others CRISPR-CAS and Thoeris tolerate and resist viral infection and replication. In mammals, viral infection activates remodeling of cellular DNA operons to initiate interferons especially gamma

isotypes to protect other normal non infected cells throughout remodeling their DNA so prevent viral integration to their genomic material ending with death of virus. CBASS ecosystems cascaded throughout activation of CD-NTase to synthesize cyclic oligonucleotides and regulation of cyclic adenosine and guanosine monophosphate (cGAS axis) to production of death associated proteins (CAPs) to execute cell death thereby inhibit phage action (Hobbs *et al.*, 2022; Wang & Zhang, 2023).

METHODOLOGY

Pacemaker Cascades 2

A primordial triple initiative trails were augmented for five stages pattern credit of biofilm charged polysaccharides slime clouds, in which verified versatile biofilm matrix diversity enrolled throughout modified roll-cup TSBYE₇₆, augmented gilded ordinary tissue culture microtiter plates assess for direct taxation of slime bounds slippery via modified chromosomal built-in Tissue Culture Assay (Microtiter plate, MTCA) cascaded by proved Congo Red media for indirect taxation of Plasmid dependent cyclones of slime armed strength. Primordial Sensitivity versus Specifically Decoration ambiguously Virulence Index of Biofilm was predictable as a confirmed mean log of together organized growth pattern protocols signified by generation time and log curve primeval design (Al-Shammary, 2009; Naves *et al.*, 2008; Dadawala *et al.*, 2010; Hassan *et al.*, 2011; Panda *et al.*, 2016; Nosrati *et al.*, 2017; Kirmusaoglu, 2019; Al-Naseri and Al-Shammary, 2020A; Fokt *et al.*, 2022; Hazaa and Al-Shammary, 2022A; Kudinha and Kong, 2022; Sanchez *et al.*, 2022; Zhou *et al.*, 2022). Excellence and Measure valuation via MTP (TCP) proved performance in which, a adapted two-fold reinforce TSBYE₇₆ was informed by a renewed active log dependent culture of McFarland (CFU.ml⁵) on a large concave wells charged negatively registered quadrilateral plate as deciphered via Christensen *et al.* (1985). Otherwise, adapted Congo red agar (Freeman *et al.*, 1989) was dependent for biofilm uncovering versatile phenotypes poured with the forecast of plasmid understanding pairs of diverse phenotypes poured clones ancestry. Al-Shammary verified *E. coli* lineage dependent Biofilm augmentation (Al-Shammary and Abdul Mounam, 2022; Al-Shammary and Abdul Mounam, 2023).

An overnight *E. coli* ancestry grown culture in dsTSBYE₇₆ at 37 °C was diluted and relocated (standardized titration) in microtiter dish as (0.5) ml almost log⁵ McFarland to 5 ml recently set dsTSBYE₇₆ decanted in separately hole. Triplicate testing recovered isolate. Therefore, imagined holes with dsTSBYE₇₆ were set as a negative control vs positive control primed Human isolate. The dishes were gestated for 48 h at 37 °C in order to edifice approval of noticeable clear biofilm cement pluri-mucoid constructions, spots and layers deposited privileged marginal rims and in the hollow of stimulating pits. Besides, the culture was aloof and plates were laved 3 times with subtle phosphate-buffered saline to eliminate non-adherent lockups and dried in an upturned situation. Devotee biofilm was fixed with 2 % sodium acetate for 5 minutes then stained with dual adapted 20 % biofilm-crystal violet and 20 % biofilm-safranin for (15-30) minutes contingent on primordial power of clone lineage, sympathy, and issue for color sort. Formerly, liberated stain was detached and the shafts were laved 3 times with PBS. Settling plates (2-3) hours for aridity then discolored spots and layers of biofilm in hollow versus everywhere interior rims of shafts were snapped, restrained and recorded rendering to the formation degree, stain diversity and isolate ancestry. Understandable consequence exposed after limited hours to day after ample aridity of persuaded biofilm. Optical density (OD) of discolored deposited biofilm attained by primed “micro-ELISA auto reader” at wavelength (OD 570.

600) nm or actual time struggle-built lockup analyzer, biosensors, fluorescent versus scanning electron microscopy. Cut-off morals for manufacture biofilm can be considered rendering to prove approaches (Stepanovic *et al.*, 2007; Gutierrez *et al.*, 2016; Larimer *et al.*, 2016; Hashem *et al.*, 2017; Kirmusaoglu, 2019). Freeman *et al.*, 1989 had deciphered a substitute scheme for broadcast biofilm establishment; which necessitates the custom of a topic designed media. A adjustment was finished by trading BHI agar via dual reinforce dsTSBYE₇₆ (10 g Tryptone Soya Agar + 1 g Yeast Extract \ 100 ml d. w.) augmented with 5% sucrose (5 gm \ 100 ml) and Congo red (10 gm \ L) for improved consequences. Congo red decanted directly with media or equipped as rigorous solution aqueous cascaded autoclaved at 121°C for 15 minutes, discretely from other constituents' medium, then decanted cooled agar to 55°C. Critical step, just boiling don't autoclave media. Inoculated plates incubated for 24 to 48 hr. at 37°C. Optimistic outcome indicated by a cyclone's growth pattern of plasmids amplified dark colonies with a crystalline dry evenness. Feeble slime producers frequently endured pink, yet infrequent darkening at the colony's centers. Colonies darkening with a dry crystalline absence versus morphology designated an midway outcome. Triplicate and recurrent 3 times intervals trial scheduled scheme.

Primordial quorum sensing, "trusted platform module" nuance via a poured network of genetically multi stress hardening amplified inside a denominator ancestry, shifting an emergent tragedy to end point of wayward views. Susceptibility antimicrobial testing (AST) for topic isolates is a shared and significant performance in utmost medical workrooms. The outcomes of these trials are cast-off for selection of the most suitable antimicrobial agent(s) redirected against the infectious denominator. Pending the 1950s, laboratories were missing policies and tackle for the precise resolve of *in vitro* replies of bugs to antimicrobial agents. Bauer *et al* (2009) instigated the expansion of standardized approaches for susceptibility antimicrobial testing, using "disc diffusion system". Yet, the vulnerability outcomes may not always correlate with the patient's retort to cure. The comeback of an infected patient to antimicrobial agent(s) is a multifaceted interrelationship of host replies, drug dynamics and microbial bustle. *in vitro* deciphered tolerant versus resistant and even persists pattern dose not confusingly or semiconservative significantly module for authenticity of action potential outline of that antibiotic *in vivo* to effort with natural defense barriers against *in vitro* resistant denominator. Susceptibility antimicrobial tests are either reckonable or topic. "Minimum Inhibitory Concentration (MIC)" willpower is a reckonable scheme. Decisive MIC using a conventional broth dilution process is a episode overwhelming. Therefore, Gradient susceptibility diffusion is widely accepted for a long time. Appropriate selection of antibiotic for handling depends on correct diagnostic signal in which, susceptibility antibiotics test (AST) settles these guidelines. Amplified resistance emergency problems of new multidrug pathogens transferred via food chain particularly in Iraqi environment drama a critical tool for hygienic community and economic strategy for manufacture and trading of safe food and customer satisfaction (Kanaan, 2013). In medical exercise, record frequently prescribed antibiotics are built on universal guidelines and information about sensitivity. Since vulnerability can vary even within a species "between and within some strains being more resistant than others are", susceptibility antibiotic testing (AST) is usually carried out to regulate which target will be utmost successful in treating *in vivo* a bacterial infection.

A semi-quantitative disc's diffusion (Kirby-Bauer outline); covering changed antibiotics levels, or soaked paper discs, are released in diverse culture zones on agar plate, which is an enriched milieu for growing bacteria. Diffused antibiotic in expanse surrounding each pill that becomes visible.

Meanwhile the concentration of antibiotic was the chief at the center, and the lowest at the edge of zone, the diameter is evocative for redirected MIC, (diameter conversion in millimeter to the MIC, in $\mu\text{g}\cdot\text{ml}^{-1}$, is constructed on notorious linear regression curves). Constructed on sanctioned supervisor skill and orders of “national committee of clinical laboratory standards (NCCLS)” formerly “clinical laboratory standards institute (CLSI, 2022)” rules tracked for Kirby-Bauer diffusion disc scheme (Bauer, 1966; Hudzicki, 2009) training the susceptibility-sensitivity of topic *E. coli* ancestry isolates to certain and gathered antibiotics (HiMedia®, India (2022): Series gradient and dose dependent discs dispersed as poured with selective pressure presented upstairs in table (3.6) were dependent for this augmented torment as in detailed manual instructions of company legislation leaflet publisher (HiMedia, 2022). Contemporary evolving performances for susceptibility antibiotic tests: Built on Syal *et al* (2017), the redirected MIC is demarcated as the lowest concentration of antibiotic vital for averting bacterial growth to regulate susceptibility index of targeted denominator. MIC does not unavoidably suggest bacterial demise, but rather growth inhibition. Consequently, MIC fluctuates from “Minimum Bactericidal Concentration (MBC)”, a suitable value which is occasionally determined in a medical laboratory due to extra exertion required. A breakpoint is demarcated as the antibiotic concentration that enables interpretation of AST to describe isolates as susceptible, intermediate, or resistant. If the recorded MIC is less than or equal to the breakpoint, then the isolate is pointed susceptible to that antibiotic.

Scored multi stress conduct was documented in multi drug resistant isolates of *E. coli* ancestry during poured urbane primordial “epigenetic tolerance drift” versus “genetic resistance shift” quorum sensing consistent machines. *in vitro* dual drive ultraviolet c susceptibility irradiation tentative perfect (Direct irradiation of inoculated TSAYE and Indirect irradiation of Pearl purified waters (by reverse osmosis with ultraviolet and ozone regimes) experimentally contaminated with five logs of *E. coli* lineage) was originated to checkup the pattern resistance of multidrug resistant of *E. coli* isolates worse from waters line (household besides purification stations) and Social source (UTI infected and carriers). The irradiation trial plan was registered in Department of Veterinary Public Health/Meat Hygiene lab for postgraduates in which, a topic multidrug resistant PCR primed genotypes of *E. coli* ancestry were recently enriched and boosted with dsTSBYE₇₆ overnight at 37 °C, then MacFarland opacity tubes titrated and standardized during numeration sequence of droplet and roll-pour plate performances to be dose dependent curve of 5 logs 10^5 CFU.ml⁻¹ for each isolate. A biosafety cabinet maintained via “15-W mercury vapor 254-nm germicidal lamp” with total emitted joint attuned dose dependent “radiation beam 25 J/cm²”. Newly cultured dishes on dsTSAYE₇₆ were exposed to these wave length at nearly 50 cm distance (critical checkpoint dishes were unlocked not covered due to UVC beam don’t penetrate covers).

Rendering to the preceding investigates in the radiology field, UVC lights kills and turns as decontamination management with this wave range length in a stated episode time intervals not exceeds 20 minutes in comparison to laser beams of infrared irradiation protocol (fifteen seconds to two minute). Positive control inoculated non irradiated cultures versus negative control irradiated non inoculated cultured dishes and contaminated pearls water (twenty-five ml of p-waters decanted in a serial disposable fifty ml cups contaminated and mixed thoroughly with one-hundred micron of freshly prepared and titrated half McFarland five logs *E. coli* lineage) were registered proved attainment. Primeval triplicates for each iconic stones culture were exposed to UVC for a long 60 minutes break in agreement to manager idea and experience. The radiation strength was intended

for inundating about an area of 15 ft² under hood. Irradiated cultures were shelters covered inside a hood then incubated at 37 °C for 24 hours. Susceptibility radiation index for selected isolates were likened to standard inhibition zones suggested antibiotics selected and standardized as a control parameter in agreement with CLSI instructions and average antibiotics tables (CLSI, 2022). All UVC light processed units with alternate pairs of control positive and negative trails were cultured directly in case of plates and indirectly from processed contaminated waters units and replicates cascaded by MM droplets and roll-tube pore plate techniques, followed by incubation at 37 C for (18-24) hours. According to irradiation strategies, a direct DNA breakdown occurred terminated with death of *E. coli mix* or tolerance vs resistance behaviors verified. Indirect hurdling with cooling processed contaminated waters units inside a refrigerator at 4 C, then repeated culturing on TSAYE upstairs either assist in termination or killing of recovered isolates mix indirectly during activation of free radicals from waters or abnormally resuscitate dormant forbidden tolerant vs resistant clones of *E. coli* lineage. Overall total, three times episodes of culturing with replicates, then recording counting results in mean logs with critics, phenotypic changing of colonies and isolates after direct and indirect impedance to UV light regimes with aid of SEM.

Ultraviolet (UV) is a formula of electromagnetic radiation with wavelength shorter than that of visible light, but longer than X-rays. The UV region covers the wavelength range “100-400” nm and is alienated into 3 bands: “UVA (315-400 nm)” “UVB (280-315 nm)” “UVC (100-280 nm)”. The devices of UVGI on microbes are exclusively susceptible to light at wavelengths at or near “253.7 nm” due to maximum absorption wavelength of a DNA molecule is “260 nm”. Later UV irradiation, the DNA sequence of microbes can initiate pyrimidine dimers, which can restrict DNA duplication, in addition to prime to devastation of nucleic acids and render the viruses noninfectious. Microbial demise persuaded by UVC has been qualified to DNA mutations, including the establishment of cyclobutyl-type dimers (pyrimidine dimers) and pyrimidine adducts. Moreover, the overproduction of reactive oxygen species (ROS) induced by UV radiation can oxidize membrane lipids and inhibit critical cellular enzymes, so Gram-negative bacteria more vulnerable than Gram-positive. “1 Gray (Gy) = 1 Joule/kilogram = 100 rad (radiation adsorption density)”. Gray degree castoff for any type of radiation (e.g., alpha, beta, neutron, gamma), but it does not decipherer the biological effects of dissimilar radiations. Biological effects of radiation are restrained in units of "sievert" (or the older designation "rem") (Diehl, 1995; Meechan and Wilson, 2006; Richa *et al.*, 2010; Cutler and Zimmerman, 2011; Falguera *et al.*, 2011; Jan, 2014; Kanval *et al.*, 2024; Vasuja and Kumar, 2018; Song *et al.*, 2019; Singh *et al.*, 2020; Vanhaelewyn *et al.*, 2020; Chacha *et al.*, 2021; Chawla *et al.*, 2021; Darre *et al.*, 2022; Yemmireddy *et al.*, 2022; Britanica, 2023).

Absolute measurement exposed the amount of UVC energy emitting power on the target per unit area, expressed in milliwatts or millijoule second per square centimeter (mW-s or mJ-s/cm²) in which, standardized inside biosafety cabinet by 40 watts mercury fluorescent lamp to a one-meter distance vertical area via exposing emitting dose dependent power 125 mw mJ.s / cm² to a cumulatively reach to 30 KGy (3 x 10⁶ rad) in about 12.5 minutes that's fair enough to disinfect the area under the hood (2 x 2 meter distance of stainless steel hood bench surrounded by special corning glass). Most susceptible microorganisms either RNA or DNA, single stranded or double stranded genetic material can be affected and killed at range between (1-10) UVC Gy. Targeted experimental module exposing these forbidden multidrug resistant recovered isolates directly on agar and indirectly within contaminated milk or water inside a biosafety cabinet for one hour period to

determining whether susceptibility patterns (tolerance vs resistance) in spite of disinfecting protocol in which, radiation dose reach approximately (4-5) folds original standardized dose (30 KGy) i.e., (120-150 KGy). Most Bacteria and viruses are sensitive to (1-10) Gy UVC for (5-15) secs.

“UV dose ($\mu\text{W}\cdot\text{s}/\text{cm}^2$) = UV intensity ($\mu\text{W}/\text{cm}^2$) \times exposure time (seconds)” (Diehl, 1995; Meechan and Wilson, 2006; Richa *et al.*, 2010; Cutler and Zimmerman, 2011; Falguera *et al.*, 2011; Jan, 2014; Vasuja and Kumar, 2018; Song *et al.*, 2019; Singh *et al.*, 2020; Vanhaelewyn *et al.*, 2020; Chacha *et al.*, 2021; Chawla *et al.*, 2021; Darre *et al.*, 2022; Yemmireddy *et al.*, 2022; Britanica, 2023).

RESULTS and DISCUSSION

Biostatistical integration was a predominant tool for deciphering and matching displayed calculated cascaded results series at confidence intervals 95 & 99 % in which, all observed results were analyzed by statistical analysis system (SAS, 2018) software program throughout interconnected values of significant and non or insignificant probability index of $p \leq 0.05$ & $p \leq 0.01$ via dependent analysis of variance (ANOVA) with least significant difference (LSD) cascaded by Chi square (χ^2) to understanding normal distribution nature of samples and their replicates. Significant values mean significant clinical observed and calculated trials of experimental design and dissertation aim & objectives. Not always non or insignificant results means they were not important clinically or scientifically in accordance to pairs of null & alternative experimental hypothesis design but this dependent primarily on virulence indices cascaded by type of evolved isolate i.e., their genetic makeup in terms of genetically modified microorganisms as priority bioterrorism special topic issue within food chain with other interconnected predisposing factors and ecosystems in terms of zone of infection cascaded by episodes times intervals. In conclusions: Clinical observed trials were very important in spite of their statistical values were not significant in some situations.

Emergent Risk Assessment Patterns illustrated and deciphered via an abnormal naturally and genetically modified, highly infectious forbidden foci derives from unknown portals of entrance or Era gates throughout cascaded bridges to be resident and deposited within Iraqi environment with continuous stress adaptation and hardening cis-trans accommodation strategies. These emergent forbidden, biohazard and bioterror *E. coli* lineage are genetically well-equipped foreign entity augmented to survive harsh environments with other infectious and contagious foci inside a recalcitrant barrier of biofilm to create an abnormal dangerous and sophisticated struggling entity with diverse and versatile, drift shift antigenic transformations ending with forbidden sequels. According to legendary futurist inventor and innovator Nikola Tesla and their AI cascaded researches in field of electromagnetic energy patterns constructions verification models with built-in forbidden sequels of negative energy and death irradiations oscillation poured by HARPA and DARPA projects for destroying every living thing and lifestyle changes, terminated with bioterrorism highly infectious agents' resident in food chain (Mustafa & Mohammed, 2014; Al-Atta, 2019; Hanoun & Al-Samrrae, 2019; Sabri & Kareem, 2020; Hamad, 2022; Salumi & Abound, 2022; Al-Shammary, 2023).

Deciphered microbial quality patterns of house tap waters, purification stations and Human UTI cases (males & females individuals & workers) unveiled abnormal complex relationships among diverse and versatile recovered isolates built-in emergent forbidden struggling (mix of pollution versus contamination versus clinical entities). Abnormal mixed phenotypic recovered entities within a biofilm including *Escherichia coli* lineage with Human denominator *EHEC serotype O157:H7*, *Staphylococcus aureus* with MRSA forbidden clone, *Pseudomonas aeruginosa*, *Proteus mirabilis* vs

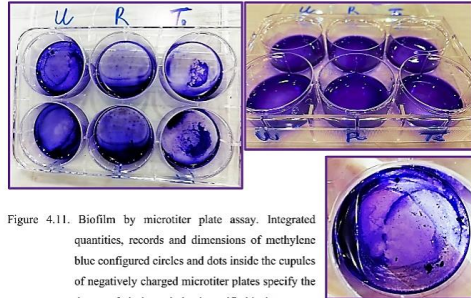
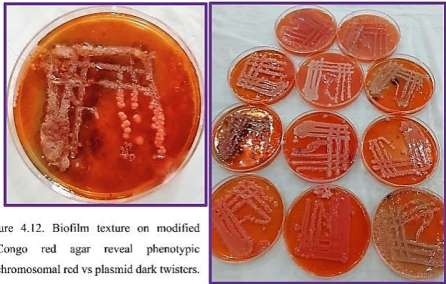
vulgaris, *Enterococcus faecalis* & *Klebsiella spp.* with forbidden pneumonic clone via HiCrome UTI Rainbow agar cascaded by HiCrome MacConkey-Sorbitol agar. These recorded phenotypic recovery ecomaps described in table 4.1. Description of colonial variants phenotypes of recovered documented *E. coli* lineage (*E. coli* & serotype *O157:H7*) on both agars revealed diverse communities of contamination-pollution series vs clinical versatile isolates. Documented and recorded clinical picture project presence of small, circular, convex with deep red to burgundy texture due to action of owned β -D-galactosidase on Chromogenic substrate. Both cis-trans *E. coli* and EHEC serotype *O157:H7* forbidden prohibited ecomaps' ancestors. Versus inhibition of contamination isolates on UTI agar (Specificity and Sensitivity Patterns). While, MacConkey-Sorbitol agar project clinical and contaminant *E. coli* sorbitol fermenter as polymorphic pink translucent colonial variants. Contaminant serotype *O157:H7* sorbitol non-fermenter as poly morphic transparent translucent phenotypes. Clinical forbidden two phenotype as transparent reverse phenotype-1 and white opaque reverse phenotype-2. These interconnected diverse and versatile colonial variants cascades described in table 4.2.

Similarity index patterns of *E. coli* lineage versus recorded upstairs foodborne and waterborne pathogens cascaded by UTI cases series were recorded and documented inside Iraqi ecosystems with diverse and versatile epidemiological segregation ecomaps (Al-Shammmary, 2009; Abdel Nafeh & Salman, 2011; Al-Shammmary & Abdul Mounam, 2011; Kanaan & Al-Shammmary, 2013; Flayyih *et al.*, 2014; Al-Shammmary, 2015a; Al-Shammmary, 2017; Al-Shammmary & Abdul Mounam, 2017; AL-Musawi *et al.*, 2018; Al-Saadi, 2019; Al-Shammmary, 2019; Abdulsattar *et al.*, 2020; Al-Naseri & Al-Shammmary, 2020a; Al-Naseri & Al-Shammmary, 2020b; Al-Shammmary & Abdul Mounam, 2020; Razooqi & Al-Shammmary, 2020a; AL-Imam & Flayyih, 2022; Al-Shammmary & Abdul Mounam, 2022; Hazaa & Al-Shammmary, 2022a; Hazaa & Al-Shammmary, 2022b; Hazaa & Al-Shammmary, 2022c; Oleiwi & Al-Dabbas, 2022; Al-Shammmary & Abdul Mounam, 2023). Special topics of this verified thesis unveiled isolation and identifications of new biohazard foci ancestor forbidden ecomap chain of an emergent epigenetically adapted *E. coli* lineage in waters chain cascaded by stress hardening (drift and shift) and quorum sensing transistors behavior. Verified data references indicate indirectly to the presence of this denominator in the waters supplies, but rather isolated from rivers, sewage, hospitals (nosocomial). But our unparalleled mission, motivation, determination, efforts, patience and courage satisfy with this augmented enrollment associated with the documentary series built-in events from recovery to molecular fingerprints arrays clearly indicate this paranoid tournament.

There are really strange and scary things happening in this world. You may not believe it, but a scary monster (a biofilm built-in entity to become a Chimera) appeared in this mess. The Chimera appears every night, and today we are gathered to decide to leave from here. What, but how and what is the form of this frightening entity, that no one has seen before, but it is scary that he runs around the village and knocks on the doors, what the wind may be, or fantasies, or something else, you mortals do not understand, it is a punishment from the great chief. Biofilm fabricators were separated rendering to the sort and grade of sliminess' via microtitre dish assess versus diversity into dark dependent plasmids cyclones and versatile red to white colonies displayed surface of confirmed Congo red agar. Entirely separated lineages recovered from sliminess' both from isolates and waters. Substantiated blue smarmy mucoid masses inside hollow in scanned topographical districts were potentiated strappingly via both schemes. DNA associated and plasmid allied cyclones were sensed in perused brands from proved topic districts with adjustable organized upshots. Dual enriched TSBYE76 parts authenticate biofilm during aggregates smarmy mucoid privileged hollow of artificial chemostat components in addition to captivating a loop from colonies with smarmy threads.

Cohesive amounts, annals and scopes of blue-red arranged rings and spots private cupules of microtiter plates negatively charged stipulate the virulence degree index in topic isolates. Indexed biofilm from isolates separate matrix into tough plasmid cyclones 'cluster including sequence entities from all districts. While, strong phenotypically chromosomal associated biofilm producers were resident from and deposited in Abu-Ghraib & Al-Sha'ab purification plans linked to those from man UTI patients, the strong plasmid twisters' phenotypes resident from and deposited in Al-Saidiya, Al-

Table 4.25. Phenotypic Chromosomal Biofilm vs Plasmid Twisters.

Codebook	Biofilm Microtiter or Tissue Culture Plate	Biofilm Texture on Modified Congo Red Agar			
		Chromosomal Red	Plasmid Twisters Black- Dark	Biofilm Non-Producer White	
CW	<p>Biofilm from both waters and recovered isolates.</p> <p>Verified blue slimy mucoid aggregates inside bottom of synthetic chemostat units as well as during taking a loop from colonies with slimy threads. Integrated quantities, records and dimensions of methylene blue configured circles and dots inside the cupules of negatively charged microtiter plates specify the degree of virulence index in verified isolates.</p>	Medium	Strong Positive	None	
GW			None		
QW			Positive	None	
AW		Strong Positive			
T1		None	White		
T2		Strong	Strong Positive	None	
T3		Medium	Positive		
T4		Medium	None		
FW			Strong Positive	None	
HW					
SW			None		
PW			Strong	Strong Positive	None
R1					
R2					
R3			Medium	None	
R4					
Ux	Strong	Strong Positive	White		

America & Al-Sha'ab household tap waters cascaded by Abu-Ghraib & Al-Sha'ab purification stations linked to those from man UTI patients. Within isolate ancestry ecosystem from the topic brand and district, existing mutable progeny with sophisticated morphological features or identical phenotype with dissimilar behaviors owing DNA and plasmid cyclones, robust and red to pink sluggish makers to white non paste producers. Downstream table (4.25.) figures (4.11. & 4.12.) illustrates and decipher outcomes.

Interconnected genes sharing strategies within recalcitrant biofilm ecosystem programed forbidden selective persists progeny behaviors with brain like machines built with an artificial intelligence cascades of quorum sensing & sigma factors orchestrated throughout forbidding conjugation bridges of transmissible plasmids twisters, equipped transformation with foreign external DNA and transduction with inserted & redirected augmented prophages to become an abnormal emergent complex polysaccharide built-in biofilm entity housekeeping behavior of Chimeras (Al-Shammary, 2023). Diverse & versatile recovered & recorded isolates from different sampled units & districts reflect plasticity of genetic materials of isolates with transmission behaviors from epigenetic transient stress adapted environment surrounding the genes as drift tolerance forbidden intermediate cascade to switching to real genetic permanent evolved mutation as shift resistance sequels (Al-Shammary, 2023). Matrixed mega & micro habitual biofilm growth patterns built-in behaviors of microbial communities reflect dangerous interconnected heterogenic plasticity in programed & regulated virulence strategies built within biofilm barrier capsule or *Mycobacterium* complex wax-D like environment & proceeds outstanding to rebuild a new struggling. Synergistic recycling of an independent & dependent ATP components within biofilm if these forbidden creatures to combat stressors with gradient stress adaptation until coordinating to stress hardening

phenomenon. Overwhelming cascaded well-orchestrated schedules that are directed through differential gene expression & regulated by molecular communication among communities. Difficult clearance situations of these enhanced specified persisters with normal regulated hygienic processing like preservation and hurdling decontamination reflect problematic emergency HACCP policy for these priority bioterrorism infectious foci (Pandeya *et al.*, 2022).

Significant diversity within family of *E. coli* lineage regulatory integrated stress adaptation genetic loci (integrons & chaperones) reflect genetic plasticity & shifting transmission during translocation from clinical case to contamination phase in which, dependent and independent genes expression & transformation sharing mechanisms occurs. Production & exaggeration of resistance genes within food chain & food processing environments via capsulated biofilm barriers represent biohazard & bioterrorism emergent sophisticated problematic struggling. In modern society, these forbidden foodborne biofilm pathogens & their contamination versus pollution impact can activate an emergent cornerstone policy (Begley & Hill, 2015).

Table 4.28. Susceptibility Patterns Index of recovered *E. coli* lineages from house tap waters in Al-Karkh zone (Nine isolates) for selected antibiotics according to split-split CLSI tables (2023).

Antibiotics	<i>E. coli</i> (2)		<i>O157:H7</i> (7)		<i>E. coli</i> lineages (9)	
	S %	R %	S %	R %	S %	R %
Aztreonam (ATM)	1	1	4	3	5	4
Imipenem (IPM)	0	2	0	7	0	9
Meropenem (MRP)	0	2	1	6	1	8
Ceftazidime (CAZ)	0	2 ESβL	0	7 ESβL	0	9 ESβL
Cefepime (CPM)	0	2	1	6	1	8
Norfloxacin (NOR)	0	2	0	7	0	9
Ciprofloxacin (CIP)	1	1	4	3	5	4
Nitrofurantoin (NIT)	0	2	0	7	0	9
Colistin (CS)	1	1	6	1	7	2
Azithromycin (AZM)	0	2	0	7	0	9
Clindamycin (CD)	0	2	1	6	1	8
Tigecycline (TGC)	0	2	0	7	0	9
Amikacin (AK)	2	0	6	1	8	1
Co-Trimoxazole (COT)	0	2	1	6	1	8
Ampicillin (AMP)	0	2	0	7	0	9
Amp/Sulbactam (A/S)	0	2	0	7	0	9

Susceptibility Patterns Index of recovered *E. coli* lineage from household tap waters in Al-Karkh sector (Nine isolates) for selected antibiotics according to split-split CLSI tables (2023). Recorded topic resistance profile (MDR, ESβL, XDR) to selected antibiotics as in table (4.26.) segregated & categorized as multidrug resistant forbidden recovered isolates except to moderate susceptibility to Aztreonam, Ciprofloxacin, Colistin, Amikacin, Amoxiclav, Augmentin & Piperacillin/Tazobactam. Table (4.27.) decipherer susceptibility patterns index of recovered *E. coli* lineage from WPP in Al-Karkh sector (Five isolates) for selected antibiotics according to split-split CLSI tables (2023). Chainsaw puzzles resistance to selected antibiotics except intermediate sensitivity to Imipenem, Norfloxacin, Ciprofloxacin, Nitrofurantoin, Colistin, Azithromycin, Clindamycin, Tigecycline (new), Amikacin, Co-Trimoxazole, Ampicillin, Amoxiclav & Piperacillin/Tazobactam. Table (4.28.) decipherer susceptibility patterns index of recovered *O157:H7* from waters in Al-Rusafa sector (Eight isolates) for selected antibiotics according to split-split CLSI tables (2023). Most recovered isolates

were susceptible to selected antibiotics except some moderate resistance to Imipenem, Meropenem, Ceftazidime (ESβL), Cefepime, Norfloxacin, Ciprofloxacin, Nitrofurantoin, Azithromycin, Tigecycline, Amikacin, Co-Trimoxazole, Ampicillin, Amp/Sulbactam, Ticarcillin/Clavulanic & Piperacillin/Tazobactam. Table (4.29.) decipherer susceptibility patterns index of recovered *E. coli* from human UTI in Baghdad Province (Sixteen isolates) for selected antibiotics according to split-split CLSI tables (2023). Recovered isolates were resistant to all selected antibiotics.

Table 4.29. Susceptibility Patterns Index of recovered *E. coli* lineages from WPS in Al-Karkh zone (Five isolates) for selected antibiotics according to split-split CLSI tables (2023).

Antibiotics	<i>E. coli</i> (1)		<i>O157:H7</i> (4)		<i>E. coli</i> lineages (5)	
	S %	R %	S %	R %	S %	R %
Aztreonam (ATM)	0	1	0	4	0	5
Imipenem (IPM)	1	0	1	3	2	3
Meropenem (MRP)	0	1	0	4	0	5
Ceftazidime (CAZ)	0	1 ESβL	0	4 ESβL	0	5 ESβL
Cefepime (CPM)	0	1	0	4	0	5
Norfloxacin (NOR)	0	1	1	3	1	4
Ciprofloxacin (CIP)	1	0	3	1	4	1
Nitrofurantoin (NIT)	1	0	4	0	5	0
Colistin (CS)	0	1	1	3	1	4
Azithromycin (AZM)	0	1	3	1	3	2
Clindamycin (CD)	0	1	1	3	1	4
Tigecycline (TGC)	0	1	1	3	1	4
Amikacin (AK)	0	1	1	3	1	4
Co-Trimoxazole (COT)	1	0	3	1	4	1
Ampicillin (AMP)	0	1	1	3	1	4
Amp/Sulbactam (A/S)	0	1	0	4	0	5
Amoxiclav (AMC)	1	0	3	1	4	1
Ticarcillin/Clavulanic acid (TCC)	0	1	0	4	0	5
Piperacillin/Tazobactam (PIT)	1	0	3	1	4	1
Total	6 (31.58)	13 (68.42)	26 (34.21)	50 (65.79)	34 (35.79)	61 (64.21)
P-value	0.108 NS		0.0059 **		0.0056 **	
** (P≤0.01), NS: Non-Significant.						

Amoxiclav (AMC)	1	1	4	3	5	4
Ticarcillin/Clavulanic acid (TCC)	0	2	0	7	0	9
Piperacillin/Tazobactam (PIT)	1	1	5	2	6	3
Total	7 (18.42)	31 (81.58)	33 (24.81)	100 (75.19)	40 (23.39)	131 (76.61)
P-value	0.0001 **		0.0001 **		0.0001 **	
** (P≤0.01).						

S = Susceptible R= Resistant

Table 4.30. Susceptibility Patterns Index of recovered *O157:H7* from waters in Al-Rusafa zone (Eight isolates) for selected antibiotics according to split-split CLSI tables (2023).

Antibiotics	Tap Waters (4)		WPS (4)		<i>O157:H7</i> (8)	
	S %	R %	S %	R %	S %	R %
Aztreonam (ATM)	4	0	0	4	4	4
Imipenem (IPM)	4	0	3	1	7	1
Meropenem (MRP)	4	0	3	1	7	1
Ceftazidime (CAZ)	4	0	0	4 ESβL	4	4
Cefepime (CPM)	4	0	0	4	4	4
Norfloxacin (NOR)	4	0	0	4	4	4
Ciprofloxacin (CIP)	4	0	3	1	7	1
Nitrofurantoin (NIT)	4	0	1	3	5	3
Colistin (CS)	4	0	0	4	4	4
Azithromycin (AZM)	4	0	0	4	4	4
Clindamycin (CD)	4	0	4	0	8	0
Tigecycline (TGC)	4	0	0	4	4	4
Amikacin (AK)	4	0	2	2	6	2
Co-Trimoxazole (COT)	4	0	1	3	5	3
Ampicillin (AMP)	4	0	0	4	4	4
Amp/Sulbactam (A/S)	4	0	0	4	4	4
Amoxiclav (AMC)	4	0	4	0	8	0
Ticarcillin/Clavulanic acid (TCC)	4	0	0	4	4	4
Piperacillin/Tazobactam (PIT)	4	0	1	3	5	3
Total	76 (100)	0 (0)	22 (28.95)	54 (71.05)	98 (64.47)	54 (35.53)
P-value	0.0001 **		0.0002 **		0.0001 **	
** (P≤0.01).						

S = Susceptible R= Resistant

Table 4.31. Susceptibility Patterns Index of recovered *E. coli* from human UTI in Baghdad Province (Sixteen isolates) for selected antibiotics according to split-split CLSI tables (2023).

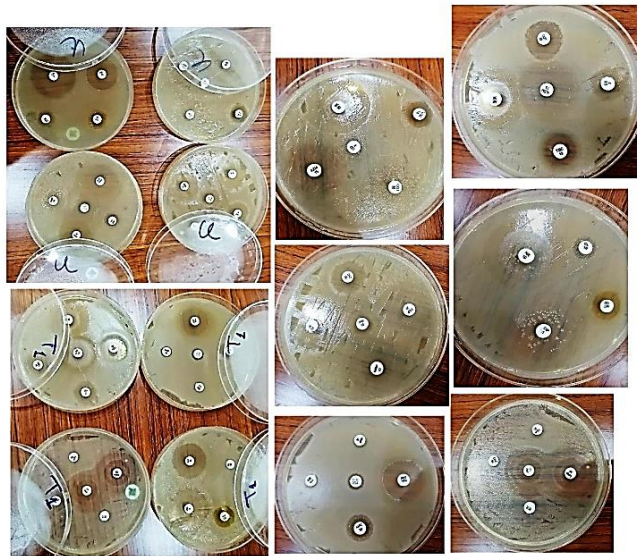
Antibiotics	Al-Karkh (8)		Al-Rusafa (8)		Baghdad (16)	
	S %	R %	S %	R %	S %	R %
Aztreonam (ATM)	0	8	0	8	0	16
Imipenem (IPM)	0	4	0	4	0	16
Meropenem (MRP)	0	4	0	4	0	16
Ceftazidime (CAZ)	0	8 ESβL	0	8 ESβL	0	16 ESβL
Cefepime (CPM)	1	7	0	8	1	15
Norfloxacin (NOR)	0	8	0	8	0	16
Ciprofloxacin (CIP)	1	7	0	8	1	15
Nitrofurantoin (NIT)	0	8	0	8	0	16
Colistin (CS)	1	7	0	8	1	15
Azithromycin (AZM)	0	8	0	8	0	16
Clindamycin (CD)	0	8	0	8	0	16
Tigecycline (TGC)	0	8	0	8	0	16
Amikacin (AK)	0	8	0	8	0	16
Co-Trimoxazole (COT)	1	7	0	8	1	15
Ampicillin (AMP)	0	8	0	8	0	16
Amp/Sulbactam (A/S)	0	8	0	8	0	16
Amoxiclav (AMC)	1	7	1	7	2	14
Ticarcillin/Clavulanic acid (TCC)	1	7	0	8	1	15
Piperacillin/Tazobactam (PIT)	1	7	1	7	2	14
Total	7 (9.21)	69 (90.79)	2 (2.63)	74 (97.37)	9 (5.92)	143 (94.08)
P-value	0.0001 **		0.0001 **		0.0001 **	
** (P≤0.01).						

S = Susceptible R= Resistant

Modified sophisticated & redirected interconnected genetic mechanisms in antibiotics resistance with epigenetic temporary tolerance drift behavior cascaded by genetic permanent resistance shift of foodborne & waterborne pathogens cascaded by genes sharing forbidden behaviors & transmission within food chain & waters ecosystems via forbidden conjugated plasmids, forbidden foreign DNA fragments transformation & forbidden CRISPR-CAS prophages transduction (Blair *et al.*, 2015; Kapoor *et al.*, 2017; Odonkor & Addo, 2018; Peterson & Kaur, 2018; Al-Shammary, 2019; Miryala & Ramaiah, 2019; Wang *et al.*, 2020; Ho *et al.*, 2021; Majumder *et al.*, 2021; Uluseker *et al.*, 2021; Wu *et al.*, 2021; Abdelwahab *et al.*, 2022; Chawla *et al.*, 2022; Elshimy, 2022; Jamrin *et al.*, 2022; Ripanda *et al.*, 2023). Uropathies UTI forbidden invaders validate extensive resistance to third-generation cephalosporins in which, clinical categorized as multiple drug-resistant (MDR) extended spectrum beta lactamase resistant (ESβL) & extensive drug resistant (XDR) (Ramchandani *et al.*,

2005; Iqbal *et al.*, 2021; Masoud *et al.*, 2022). Novel antibiotics trials combating these forbidden struggling but later & due to modified resistance strategies cascaded by emergent quorum sensing, stress adaptation & stress hardening for transmission of stress resistance by verified stimuli to ending in multiple stressors resistance even not encountered in their life cycle. Resistance mediated by interconnected diverse & versatile cascaded forbidden trails upstairs such as changing modifications in cell wall permeability, insufficient dose dependent concentrations, alterations in targeted sites, efflux pumps expulsion of the drug outside the cell, decreasing sensory metabolites, production of verified enzymes, including beta lactamases, modified aminoglycoside & chloramphenicol acetyltransferases that inactivate the antibiotics before they can exert their effects.

Overwhelming frequency & distribution patterns of mobile genetic elements a transmissible plasmid among versatile & diverse microbial population resident & deposited within forbidden recalcitrant biofilm barrier entity & food chain with transmission phases of contamination & clinical exaggeration struggling to become a forbidden CHIMERAS (Al-Shammary, 2019; Abdelwahab *et al.*, 2022; Elshimy, 2022). Primordial quorum sensing, “trusted platform module” nuance via a poured network of genetically multi stress hardening amplified inside a denominator ancestry, shifting an emergent tragedy to end point of wayward views. Combo segregation of recovered clones and their versatile phenotypes into resistant, tolerant, persisters, intermediate and susceptible entities depend on ecosystem of samples and territories guided by their broad-spectrum genetic module (interconnected drift and shift module among recovered clones and even within their phenotypes). All recovered PCR confirmed clones of *E. coli* lineage were highly resistant *in vitro* disc diffusion to selected and grouped antibiotics except some phenotypes were partially sensitive to norfloxacin, aztreonam and azithromycin with the development of authentication module of residual forbidden persisters inside inhibition zones, and an emergent evolution of dangerous verdicts of extended spectrum beta lactamase resistance (ES β L) phenomenon between and within versatile recovered phenotypes. Encapsulated slime bulldozers entities were difficult to processing with antibiotics but other side of trueness might be fault in comparison to *in vivo* coordinated ecosystem therapy (Al-Shammary, 2019; Abdelwahab *et al.*, 2022; Elshimy, 2022). Downstream figure (4.15.) illustrate & decipher matrixed antibiotics resistance of recovered *E. coli* lineage isolates. Downstream figure (4.16.) illustrate similarity index viewed & presented by VITEK® 2 antibiotics susceptibility patterns for 18 selected & spectrophotometer read antibiotics comb segregates.



Diverse & versatile profile index of *E. coli* lineage recovered from waters & UTI patients in Baghdad for antibiotics susceptibility by manual disc diffusion method compared to VITEK® 2 protocol in

which, sensitivity & specificity of recovered isolated to selected antibiotics by VITEK® 2 not reflect real complexity patterns of susceptible isolates to resistance phase in comparison to real manual complementary test, therefore actual reading depends on both parameters. Colloquially, minimum inhibitory concentrations (MICs) were calculated for selected antibiotics via VITEK® 2 in order to determine maximum bactericidal concentrations (MBCs) during period of cascaded processing or therapy regime.

Susceptibility Information		Card: AST-N222	Lot Number: 6221819203	Expires: Nov 19, 2022 12:00 CST	
		Status: Final	Analysis Time: 7.55 hours	Completed: Mar 28, 2022 21:00 CDT	
Antimicrobial	MIC	Interpretation	Antimicrobial	MIC	Interpretation
Ticarcillin	>= 128	R	Amikacin	<= 2	S
Ticarcillin/Clavulanic Acid	<= 8	S	Gentamicin	<= 1	S
Piperacillin	>= 128	R	Tobramycin	<= 1	S
Piperacillin/Tazobactam	<= 4	S	Ciprofloxacin	<= 0.25	S
Ceftazidime	<= 1	S	Pefloxacin		
Cefepime	<= 1	S	Minocycline	<= 1	S
Aztreonam	2	S	Colistin		
Imipenem	<= 0.25	S	Rifampicin		
Meropenem	<= 0.25	S	Trimethoprim/ Sulfamethoxazole	<= 20	S

Figure 4.16. Antibiotics susceptibility patterns of *E. coli* lineages recovered from waters & UTI patients in Baghdad by VITEK® 2. Adopted from ASCO center (Harthiya, Baghdad).

Catastrophic forbidden ultraviolet tolerance cascaded by resistance behaviors were observed in exposed selected MDR *E. coli lineage* recovered from waters & UTI patients during experimental processing with cold pasteurization ultraviolet scheduled regimes (within specified time & temperature as decimal log reduction time curve D-values) directly of cultured contaminated plates with five logs McFarland titer CFU.ml⁻¹ & an indirect induced contaminated waters with cooling refrigeration episodes. Biohazard & bioterror *E. coli* lineage were predominant, resident & deposited from Al-Amil house tap waters, Al-Saidiya & Al-Sha'ab purification stations cascade by Human UTI recovered isolates with violation emergency. Downstream table (4.32) & figure (4.17.) illustrate these observed forbidden sophisticated sequels.

Table 4.32. Ultraviolet Irradiation Susceptibility Index with D-values for selected multidrug resistant (MDR) *E. coli mix*: (13 isolates). Split-Split table cascades.

Selected MDR	Mean Reduction Log Count CFU.ml ⁻¹ after Ultraviolet Irradiation					
	Log	D ₀	Log	ID ₁	Log	ID ₂
AW	2	0.33	2	0.33	0	0.2
CW1	2	0.33	0	0.2	0	0.2
CW2	0	0.2	0	0.2	0	0.2
QW1	4	1	0	0.2	2	0.33
QW2	4	1	2	0.33	0	0.2
GW	0	0.2	2	0.33	0	0.2
T1a	2	0.33	5*	1.2	5*	1.2
T1b	2	0.33	5*	1.2	5*	1.2
T2	2	0.33	0	0.2	0	0.2
T3	3	0.5	0	0.2	0	0.2
HW2	2	0.33	2	0.33	0	0.2
R2	2	0.33	4*	1	1	0.25
UX	0	0.2	0	0.2	1	0.25

(UVC dose dependent 120-150 KGy)

- * Phenotypic Colonial Variants Abnormal Mega Biofilm Defense Producers.
- > Reciprocal Titrated MacFarland Pattern Fivefold Contamination Log (10⁵ CFU.unit⁻¹).
- > Ultraviolet Exposing Period 60 minutes.
- > D (Decimal) value = (T2-T1) / (Log N1-Log N2) (Adams & Moss, 2008).
- > D₀ = Direct Survival Growth Pattern of UV-processed cultured plates at 37 °C for 24 hours.
- > ID₁ = Indirect Survival Growth Pattern of UV-processed contaminated waters units at 37 °C for 24 hours.
- > ID₂ = Indirect Survival Growth Pattern of resuscitated UV-processed contaminated waters units at 4 °C for 24 hours.
- > Tolerant Z values ≥ Zero Score Versus Susceptible Z values ≤ Zero Score.
- > D-value (microbiology) - the decimal reduction time, the time required at a certain temperature to kill 90% of the organisms being targeted.
- > Z-value (microbiology) - the decimal reduction temperature, the temperature required at a certain time to kill 90% of the organisms being targeted.



Figure 4.17. Prohibited Chimeras *E. coli* lineages within biofilm recovered on TSAYE from waters & UTI cases in Baghdad especially Al-Saidiya Chimera.

Diverse & versatile stress adaptation for ultraviolet radiation consult encountered within food & waters chains ecosystems for bioterrorism pathogens, viruses like Corona, fungi, spores cascaded by forbidden biofilm entity (Khan *et al.*, 2022). UV light emissions could kill microbes directly throughout break down of DNA or indirectly throughout oxidative stress via releasing & activation of free radicles. Genetically evolved biofilm microorganism could modulate UV irradiation struggling damage of DNA via cleaver buffering response throughout production of specific shock proteins such as catalase, superoxide dismutase & endonuclease IV, which repair oxidative damage (Linden & Malley, 2002; Begley & Hill, 2015; Al-Shammery & Abdul Mounam, 2022; Hazaa & Al-Shammery, 2022c). cytotoxic B wavelength of UV light irradiation could either terminates forbidden microbes or convert them to a Chimeras like the predominant mother prokaryotic *Deinococcus radiodurans* catastrophic polyextremophilic creature that transmit evolution of radiation tolerance to other cross sectional foodborne & waterborne bacteria (Linden & Malley, 2002; Kumar *et al.*, 2021). Resistance behavior to irradiation could developed from resistance of microbes & their genes to antibiotics (Destiani & Templeton, 2019; Al-Shammery & Abdul Mounam, 2022; Hazaa & Al-Shammery, 2022c). Repeated water disinfection processing with traditional chemicals like Perchlorate rather than irradiation & ozonation cascaded by filtration residues could lead to evolution & development of tolerance & resistance problems to most if not all sublethal stressors (Grandío *et al.*, 2020; Daer *et al.*, 2022). Releasing patterns of biohazard radionuclide created via nuclear & civil industries within society cascaded by forbidden impact on Human, animals & environment. Frequency & distribution patterns of these radiation elements within polluted & contaminated environments could activate hidden uncounted sensation stimuli & stress adaptation cascaded by stress hardening via pathogenic microbes to become epigenetic temporary drift tolerance behavior or switched to genetic permanent mutants shifted resistant Chimeras with overproduction & recreation of recalcitrant photoprotective barrier of biofilm entity. Harbored buffering & anti-irradiation strategies including biosorption, biotransformation, biomineralization and intracellular accumulation (Linden & Malley, 2002;

Goldman & Travisano, 2011; Fernandez *et al.*, 2020; Al-Shammary & Abdul Mounam, 2022; Hazaa & Al-Shammary, 2022c).

Genomic diversity & plasticity (remodeling & evolutionary) in epigenetic tolerance or genetic resistance inside *E. coli* depends primarily on growth phase patterns of targeted denominator upon switching from environmental contamination or pollution phase in waters or food chain ecosystems to clinical phase in patients as UTI or foodborne illness (food poisoning). Residence & deposition of radiation repair mechanisms were extremely different in both lifestyle of invading species. Generation time of denominator (growth patterns from lag phase proceeds to log followed by exponential or stationary ending with decline primordial curve to death with intermittent early, mid & late growth evolutionary) cascaded by type & source of UV light emission processing could segregate genetic ability & capacity to tolerate irradiation shock according to selective pressure fortunately in some not all evolved forbidden clones. Repeated exposure to ultraviolet natural radiation cascaded by induced irradiation regimes causing memory shifting from exponential resistance to lag tolerance & so on new progeny specially those expressed as persisters inside a biofilm could create abnormal mutant tolerant ancestor (Linden & Malley, 2002; Selveshwari *et al.*, 2021; Al-Shammary & Abdul Mounam, 2022; Hazaa & Al-Shammary, 2022c). Different functional groups were evolved within mutated populations of *E. coli* during exposure time & temperature of cold ultraviolet pasteurization regimes in different locational structures of cell members integrity and housekeeping proteins & DNA polymerases. These elicited events in signal transduction, transcriptional & translational shifting phases of phenotyping altered growth patterns of targeted denominator *E. coli* causing sophisticated struggling hygienic biohazard problems both in Human & Environment industries. Therefore, genomic signatures of these evolved, emergent, forbidden, & adapted mutants infectious biohazard foci could not be terminated if there is no activated verified HACCP policy torment (Linden & Malley, 2002; Selveshwari *et al.*, 2021; Al-Shammary & Abdul Mounam, 2022; Hazaa & Al-Shammary, 2022c). Drifted versus drifted bridges of *E. coli* lineages in tap drinking waters and UTI cases could be combat by divers and versatile modules (Baquer, 2013; Ewaid *et al.*, 2018; Abed *et al.*, 2019; Al-Ansari *et al.*, 2019; Al-Atta, 2019; ; Al-Oqaidy & Al-Oqaidy, 2019; AL-Nasrawi & AL-Hashimy, 2020; Abdelrahman *et al.*, 2021; Fazaa *et al.*, 2021; Ghafil *et al.*, 2022; Al-Dabbagh, 2022; Abed Al-Azzawi & Abdullah, 2023).

Documentary records data unveiled multi stress patterns of recovered isolates to versatile antibiotics with shifting diversity to tolerate irradiation pasteurization. In conclusions: Contamination of tap waters with multi stress hardening *E. coli* mix cascaded with similarity genetic indices and phylogenetic tree ancestral relationships with clinical isolates from Human UTI cases as a forbidden complex sequel.

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