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Emerging Carbon-Based Biosensors & Their Rapid Diagnosis for Various Types of Cancer – Current Status and Future Prospects

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Abstract: Over the last three decades, biosensors have undergone extensive evolution; meanwhile, studies in this domain have gained widespread acceptance over the past fifteen years. Nanotechnology is being used to develop theragnostic nanomedicines for cancer diagnosis and therapy due to the distinctive physicochemical characteristics of materials at the nanoscale. Experts strive towards developing new methodologies for cancer therapy and **diagnostics** as fresh advances emerge accessible. Early ailment detection strengthens the odds of recovery and lowers the cost of treatment. Developing rapid, precise, and credible biosensing devices proves essential in boosting human life and well-being. Carbon-based nanoparticles have garnered considerable curiosity for their exceptional efficacy in biosensors. The current overview provides a meticulous summary of the most recent breakthroughs in carbon-based biosensors.

Keywords: biosensors, carbon-based biosensors, cancer diagnostics, biomarkers, **carbon dots**, carbon nanotubes, graphene

1. INTRODUCTION

Cancer Across Borders – A Global Concern: Cancer is a global epidemic causing immense expense to society [1], as documented by the Indian Council of Medical Research-National Cancer Registry Programme**,** indicating an escalating incidence in states across India. The American Cancer Society predicts that the worldwide cancer epidemic will soar by 21.7 million new diagnoses before the end of 2030 [2]. Cancer can take nearly 200 different colours, including lung, prostate, ovarian, breast, hematologic, skin, colon, and leukaemia. Environmental triggers like smoke from cigarettes, alcoholic beverages, exposure to radiation, and chemicals, as well as genetic causes like genetic mutations and autoimmune diseases, have been linked to a higher chance of cancer diagnosis [3]. Chemotherapeutics face challenges like ineffectiveness and poor drug delivery, necessitating significant doses for therapeutic outcomes. The rise in clinical examinations emphasizes the need for specific, swift, and reasonable analyses. Nano-biosensors have helped differentiate analytes and provide extensive information about biomolecular characteristics for various ailments.

2. MATERIALS AND METHODS

2.1. Biosensors

Per the World Health Organisation, biomarkers are referred to as any kind of substance, arrangement, or mechanism that can be observed within the body or its results and contribute to or forecast an occurrence of an event or disorder [4]. A sensor is the core component in making input variables into measurable signals. In his debut article, the pioneering inventor of biosensors, Leland Charles Clark **Jr,** outlined all the components that make up a biosensor. This 1956 paper dealt with a polarographic oxygen electrode, which determines blood's levels of oxygen concentration [5]. Biosensors are versatile integrated receptor-transducer devices which link together a receptor (biochemical recognition system) and a detector (transducer) capable of transforming the biochemical (biological) response into an accurately measured output signal [6]. Figure 1 represents a schematic depiction of a biosensor. In light of their knack for identifying disease-related biomarkers promptly, sensitively, label-free, affordably, and in real-time, biosensors are considered to be one of the most optimistic technologies [7]. **Moreover, n**anomaterials (NMs) are potential candidates used in the fabrication of biosensors.

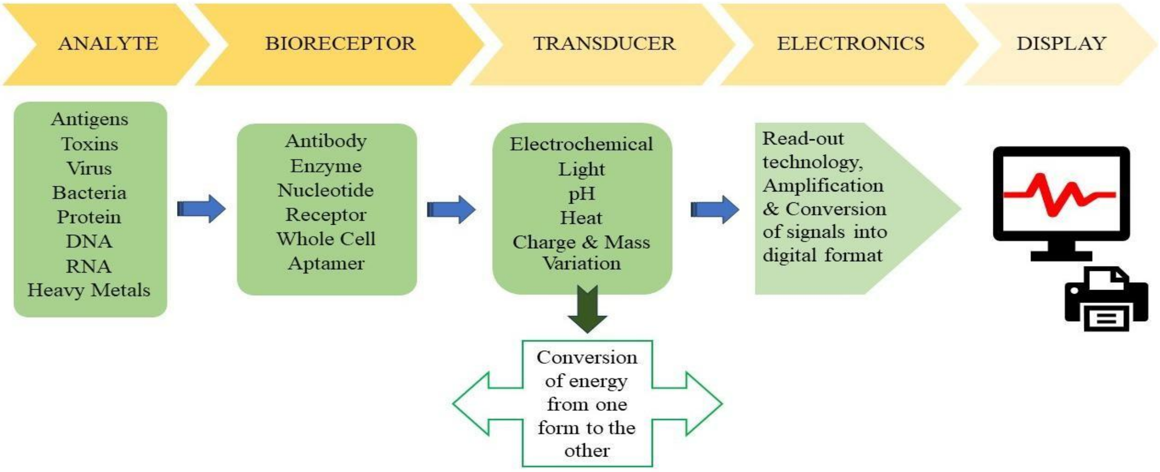

Figure 1: Schematic Representation of a Biosensor

The realm in the field of nanotechnology-based cancer therapeutics has accelerated in recent times, with an extensive variety of carbon-based nanomaterials being researched as drug delivery agents, and this success is attributed to their phenomenal thermal, electronic, and mechanical properties, alongside their elevated drug-loading capacity.

2.1.1. Types of Biosensors: Biosensors have been divided as enzymatic (the more prevalent), immunosensors (beneficial for diagnosis), aptamer/nucleic acid-based (specific for microbial organisms and nucleic acid-containing analytic substances**),** and microbial/whole-cell biosensors [8]. Biochemical reactions come into play while picking transducers. Hence, biosensors are majorly classified into electrochemical, optical, thermal, piezoelectric, etc. Electrochemical biosensors detect chemical changes in biomolecules, while optical biosensors detect light characteristics, thermal biosensors detect temperature, and piezoelectric biosensors detect mass changes [9]. Under entirety, the classification of biosensors is given in **Figure 2**.

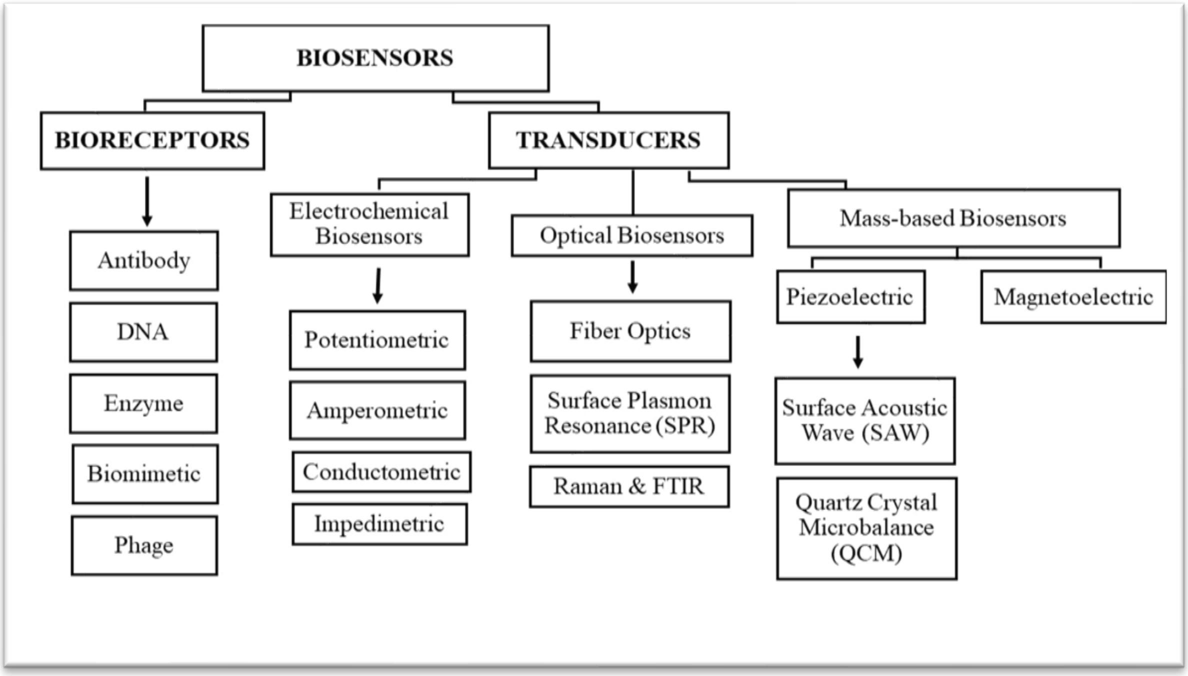

Figure 2. Classification of Biosensors

2.1.2. Role of biosensors in cancer detection: Cancer exhibits a significantly elevated mortality rate in comparison to other illnesses. It is an outbreak wherein abnormal cells develop untamed and spread across various body parts, impacting healthy cells [10], causing the treatment to be quite a tragic procedure. The expense of cancer therapy is enormous for the average individual. Biosensors' versatile nature and effectiveness lend themselves to being highly beneficial in cancer diagnosis, where early and precise detection counts for successful therapy and better treatment results for patients. The identification of cancer biomarkers is thus a crucial way to locate the onset sites of cancer cells to treat cancer in its infancy [11]. In recent years, researchers have focused on developing biosensors for an assortment of cancer categorizations, including cases of breast cancer, prostate cancer, lung cancer, ovarian cancer, colorectal cancer, and a few more. Table 1 schemes out the cancers and their relevant sensing mechanisms and biomarkers.

Table 1. Types of cancers and their relevant sensing mechanism and biomarkers

Type of Cancer	Sensing Mechanism	Biomarker Detected	Reference
Breast Cancer	Electrochemical Biosensor	HER2, CA 15-3, CA 27.29	[12]
Prostate Cancer	Optical Biosensor	PSA (Prostate-Specific Antigen)	[13]
Lung cancer	Nanomaterial-based Biosensor	CEA (Carcinoembryonic Antigen)	[14]
Colorectal Cancer	Aptamer-based Biosensor	CEA, CA 19-9	[15]
Ovarian Cancer	Electrochemical Biosensor	CA 125	[16]
Pancreatic Cancer	Colorimetric Immunosensor	CA 19-9, CEA	[17]
Liver Cancer	Electrochemical Immunosensor	AFP (Alpha-Fetoprotein)	[18]
Bladder Cancer	Fluorescent immunosensor	NMP22 (Nuclear Matrix Protein 22)	[19]
Cervical Cancer	Microfluidic Biosensor	HPV DNA	[20]
Gastric Cancer	Electrochemical immunosensor	CA 72-4, CEA	[21]

3. REVIEW ON THE DEPENDENCE OF CARBON-DERIVED BIOSENSORS IN CANCER RESEARCH

Carbon-derived nanomaterials, including various dimensions and hybridization types, have sparked interest in academia. Other forms of carbon with particular dimensions (0D, 1D, 2D, 3D) and hybridisation types (sp, sp^2, and sp^3) have been relied on [22].

The advent of sophisticated carbon-based biosensors, comprising carbon dots, carbon nanotubes, graphene, graphene oxide, and reduced graphene oxide, opens up exciting prospects for cancer detection. Figure 3 gives the classification of the carbon-based nanomaterials. These materials have high sensitivity, specificity, and adaptability, making them useful for early-stage cancer diagnosis.

Hybridization with polymers, metal oxide nanoparticles, and metal nanoparticles enhances their implementation and introduces binding sites for biomolecules or antibodies. Table 2 gives a detailed review of the literature on carbon-based biosensors in the field of oncology [23.24].

Table 2: A short Literature Review on **Carbon dots**-based biosensors in oncology

Carbon - based NMs	Probe	Matrix	Sensing mechanism	Type of cancer	Analyte	LOD	Ref
CARBON DOTS (CDs)	CD Aptamer	PAMAM - Dendrimers/ Gold NPs	Immunosensor	Ovarian cancer	Ovarian cancer biomarker CA 125	0.5 fg/mL	[25]
	CD - Nucleic Acids	Antibodies conjugated Oligonucleotide initiator	Fluorescence Sensor	Liver Cancer	Alpha Fetal Protein (AFP)	94.3 fg/mL	[26]
	CD - Antibody	Polyclonal anti-BSA capped gold NPs with FITC-labelled monoclonal anti-BSA	Immunoassay	Liver Cancer	Alpha-L-fucosidase (AFU)	3.4 nM	[27]
	CA15-3 antibody - CDs	Gold NPs - PAMAM - Aptamer	Immunosensor	Breast Cancer	Breast cancer biomarker CA 15-3	0.9 µU/mL	[28]
	CDs and Graphene oxide (GO)	Aptamer – CDs, Aptamer – CDs/GO	Aptasensor	Breast, Ovarian, Lung and Pancreatic cancers	Mucin 1 peptide	17.1 nM	[29]

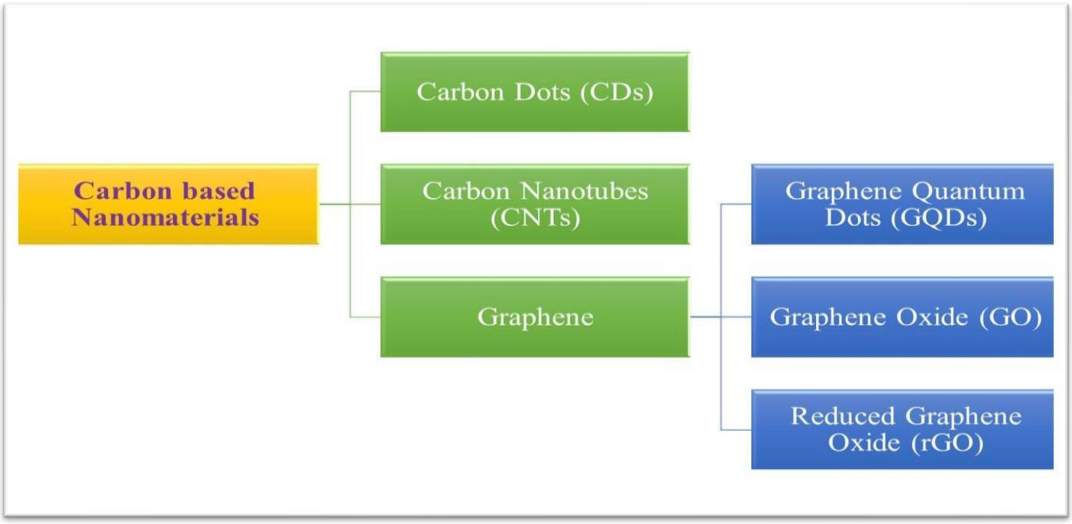

Figure 3: Classification of Carbon-based Nanomaterials

3.1. Carbon Dots (CDs): Carbon dots constitute carbon, oxygen, hydrogen, and nitrogen atoms, with varying ratios amongst unprocessed and synthesized specimens. The chemistry underlying the breakdown of crude candle black soot (91.7% C, 1.8% H, 1.8% N, 4.4% O) differs profoundly from pure CDs (36.8% C, 5.9% H, 9.6% N, 44.7% O) [23]. Carbon dots, small fluorescent nanoparticles, are widely used for imaging, visualising cancer cells, and identifying molecular biomarkers, making them valuable for early cancer diagnosis. CDs are widely used as probes for analyzing analytes in biological and environmental systems due to their size, biocompatibility, functionality, low cytotoxicity, fluorescence, and catalytic characteristics [24].

Hamd-Ghadareh and colleagues [25] devised an ultrasensitive FRET-based immunosensor capable of detecting the CA125 tumor marker and ovarian cancer cells at a dose of 0.5 fg/mL. Xu et al. [26] implemented a dual amplification fluorescence sensor having a lower detection ultimatum of 94.3 fg/mL for detecting alpha-fetoprotein, a tumor marker linked to liver cancer, using an immuno-hybridization chain reaction and metal-enhanced fluorescence. Mintz *et al*. [27] developed a biosensor for early liver cancer screening using carbon dots and gold nanoparticles. The assay, featuring an admissible limit of 3.4 nM, detects alpha-L-fucosidase in human blood, achieving high selectivity and sensitivity, and works in the PBS buffer and serum.

Mohammadi and his colleagues [28] formulated an efficacious immunosensor targeting breast tumor biomarker CA15-3, involving antibody-functionalized carbon dots and gold nanoparticles linked with a dendrimer and aptamer, thereby outperforming standard ELISA tests in sensitivity and cost-effectiveness. In the research work of Jyoti Korram and other researchers [24], a bimodal detection system was devised using S-doped carbon dots (OCDs) to retrieve prostate-specific antigen (PSA), a kind of biomarker for cases of prostate tumors. The system showed a broader linear range and low detection value, effectively detecting PSA in human blood serum.

Ding, Ling, and colleagues [29] have developed an ultrasensitive aptasensor for detecting Mucin 1 protein using FRET amid carbon dots and graphene oxide, with a threshold of 17.1 nM. Alarfaj and teammates [30] initiated a fluorescent immunoassay to analyze CA 19-9, a pancreatic cancer biomarker, in human blood samples, making use of a carbon quantum dots/gold nanocomposite material, offering excellent selectivity, accuracy and a low detection value of 0.007 U/mL. Xia's group [31] developed a ratiometric fluorescent bio probe for detecting exosomal microRNA-21 using carbon dots and DSA, achieving excellent sensitivity and a detection limit of 3.0 fM.

3.2. Carbon Nanotubes (CNTs): Carbon nanotubes (CNTs) are crucial in cancer detection by virtue of their resilient conductivity to electricity, rigidity, and extensive surface area. They are ideal for sandwich-type biosensor processing due to their stability, minimal background noise, and biocompatibility [32]. CNTs can be functionalized with bioreceptors to detect small concentrations of cancer biomarkers in biological solutions, increasing diagnosis precision.

Xin Yu and colleagues [33] developed a method to monitor tumor biomarkers like prostate-specific antigen using SWNT immunosensors and Ab2-CNT-HRP bioconjugates. The method achieved a detection bound of 4 pg mL^{-1}, surpassing commercial immunoassays and indicating potential for clinical cancer biomarker screening. Jin and cohorts [34] developed a low-cost, responsive MWCNTs@paper biosensor for detecting CA19-9, a biomarker for early-stage pancreatic cancer, with a linear concentration-resistance response.

Anshu Thapa and others [35] created a sensitive electrochemical impudent biosensor with an in-layer architecture of polyethyleneimine and carbon nanotubes, demonstrating chemisorption of CA19-9 antigen and immobilised antibodies. Sungkyung Ji's team [36] created a cost-effective, sensitive, and simple paper-based biosensor for prostate cancer detection, using bioactivated MWCNTs and micro-pore filter paper. The sensor can detect PSA levels from 0 to 500 ng/mL within 2 hours, with a 1.18 ng/mL detection edge value. Juliana C. Soares and her team [37] developed sensitive and selective immunosensors using electrospun nanofibers coated with MWCNTs or gold nanoparticles. These sensors with detection limits of 1.84 and 1.57 U/mL for MWCNT and AuNP-based systems, respectively, can detect human serum from pancreatic cancer patients with high CA19-9 concentrations.

Makableh et al. [38] developed an aptasensor for the rapid, definitive finding of human epidermal growth factor receptor 2 (HER2), a novel biomarker in breast cancer diagnosis. The sensor responded in 5 minutes and had an apprehension margin of 4.4 Pg/ml, making it an efficient early diagnosis tool. Majd et al. [39] created an n-type FET platform for detecting the cancer marker protein CA 125 in human blood samples using flexible PMMA substrates and high surface area rGO nanosheets. The FET-type aptasensor showed good sensitivity and selectivity, with a detection threshold of 5×10^{-10} U/mL. The research group of Chen [40] engineered a new electrochemical biosensor for detecting MALAT1, a potential biomarker for tracking non-small cell lung cancer. The sensor has a small detection threshold of 42.8 fM.

3.3. Graphene & Its Derivatives: Graphene, a two-dimensional carbon layer, is ideal for biosensor applications due to its electrical and mechanical properties [41], including its large surface area and conductivity. Its flexibility allows for wearable and implanted sensors, and its ability to combine carbon-based materials enhances its integration.

Jang et al. [42] developed a 3D prostate-specific antigen immunosensor using graphene-gold composites for reliable tumor diagnosis with a low detection limit of 0.59 ng/mL, while Hao and the team [43] developed a highly sensitive aptametric nanosensor for the lung cancer biomarker IL-6, with a low detection ultimatum of 139 fM. Chammari Pothipor's team [44] developed a responsive electrochemical biosensor for detecting microRNA-21, a breast cancer biomarker, integrating a screen-printed carbon electrode alongside gold nanoparticles. The cost-effective, label-free sensor, having a low detection edge of 0.020 fM, has the potential for early detection. Del Real Mata et al. [45] developed a nanostructured microfluidic system for plasmon-assisted electrochemical determination of hydrogen peroxide, a chemical produced by malignant cells, using gold nanocavities and graphene nanosheets.

Lin and his aides [46] created a genosensor to identify exon-19 mutations in the EGFR gene linked to non-small cell lung carcinoma. The sensor, made of a screen-printed carbon electrode and a pyrene-functionalized zirconia-graphene nanocomposite, demonstrated high specificity, repeatability, and stability after 8 days with a threshold for identification of 1.7 nM. Fang et al. [47] used graphene and starch to detect neuron-specific enolase (NSE). The sensor has a limit of detection (LOD) value of 0.008 pg/ml for NSE.

Graphene quantum dots (GQDs) are diminutive carbon nanomaterials with a margin particle size of less than one hundred nm. Ashish Kalkal and his colleagues [48] put together a fluorescent biosensor for ultrasensitive lung cancer biomarker screening using graphene quantum dots and gold nanoparticles, demonstrating high cell viability, quenching efficiency, and energy transfer efficiency with an average recovery of 94.69%. Weidan Na and colleagues [49] developed a fluorescent biosensor for detecting acid phosphatase (ACP) using FRET, enabling scanning of human prostate malignant cells with a minimal detection edge of 28 $\mu U\ mL^{-1}$. Fangchao Cui and his research team [50] fabricated a magnetic fluorescence biosensor to ensure prompt and sensitive isolation and detection of circulating cancer cells in the entire blood stream using graphene quantum dots conjugated with an aptamer targeting EpCAM receptors, with minimal cytotoxicity and high capture efficiency and a decreased detection limit (LOD) of 1.19 nM for EpCAM. Nian-Lu Li and his team's [51] biosensor, featuring a nanocomposite of polydopamine and silver nanoparticles for primary antibody attachment and carbon quantum dots and gold nanoparticles for secondary antibodies, has a detection edge of 1.67 pg/ml. Patrick Vilela's [52] experiment using graphene oxide as an optical sensor and upconversion nanoparticles for detecting mRNA biomarkers related to Alzheimer's Syndrome and prostate cancer demonstrated high efficiency and selectivity. Tabar and his team [53] developed a graphene-based biosensor for prostate cancer detection using PSA levels, achieving a low detection bound of 1.67 pg/ml. Shuai and his team [54] designed a sensitive sandwich-type microRNA biosensor using MgO nanoflowers and graphene oxide-gold nanoparticle hybrids, detecting miRNA-21 with high specificity and sensitivity. Deepa et al. [55] developed a CD59 immunosensor using a graphite electrode with graphene oxide functionalization, demonstrating exceptional electrochemical performance in quantifying CD59 concentrations that vary between 1 fg/ml and 10 ng/ml.

Reduced graphene oxide (rGO), a flexible material with high electrical conductivity and biomolecule binding, is being used in early cancer diagnosis due to its high sensitivity and specificity. Shawky & El-Tohamy [56] developed an ultrasensitive electrochemical immunosensor using silver/titanium dioxide/rGO nanocomposites to detect breast tumor antigen CA 15-3 in human blood samples, comprising a threshold for detection at 0.07 UmL^{-1}. Shine Augustine and colleagues [57] developed an ultrasensitive biosensor for HER-2, demonstrating enhanced sensitivity and a low detection limit. Jozghorbani and colleagues [58] constructed an electrochemical sensor for hasty disclosure of CEA using an rGO-enhanced electrode, offering a low detection limit and a large surface area. Joshi and companions [59] developed a microcontroller-based graphene biosensor to detect CYFRA 21-1 and CEA, demonstrating the importance of mobility in biosensor design and its ability to transfer charges. The sensor detected CYFRA 21-1 and CEA with an ultimatum of 0.04 pg/ml and 0.148 pg/ml, respectively.

CONCLUSION

Carbon-based biosensors have the potential to revolutionize cancer detection and oversight. These biosensors, including graphene, carbon nanotubes, and fullerenes, can detect cancer impulses **in** small amounts, enabling rapid medical care and diagnosis. Their unique electrical, thermal, and mechanical characteristics make them highly responsive. Integration with new technologies like microfluidics and wearable devices can enable portable, real-time monitoring systems for early diagnosis and ongoing cancer surveillance.

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