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Tumor microenvironment and the role of artificial intelligence for breast cancer detection and prognosis – a mini-review

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Abstract

A critical gap of knowledge has been noted in breast cancer detection, prognosis, and evaluation between tumor microenvironment and associated neoplasm.

Artificial intelligence has multiple subsets or methods for data extraction and evaluation, including artificial neural networking, which allows computational foundations, similar to neurons, to make connections and new neural pathways during data set training. Deep machine learning and artificial intelligence hold great potential to accurately assess Tumour Micro Environment (TME) models employing vast data management techniques.[1-6]

Despite the significant potential AI holds, there is still much debate surrounding the appropriate and ethical curation of medical data from Picture Archiving and Communication Systems (PACS). Artificial Intelligence (AI) output's clinical significance holds its outcome based on its human predecessor's data training sets. Integration between biomarkers, risk factors, and imaging data will allow the best predictor models for patient-based outcomes.

Introduction:

The tumor microenvironment or surrounding stroma contains various vital components such as immune cells and extracellular matrix, which act against antitumor immune cells. This leads to tumor progression, and ultimately metastasis[7-10]. The stromal environment contains many interesting signaling pathways and molecular structures related to prognostic outcomes of breast cancer[9]. The genetic alterations of cancer cells related to signaling pathways control both the processes of tumorigenesis and progression. These alterations are due to overexpression of oncogenic mutations such as growth factor receptor tyrosine kinases and nuclear receptors such as estrogen receptors. Due to the above complexities related to cancer signaling networks, the efforts to produce anticancer drugs are challenging due to inordinate signaling pathways, translating to pathway reactivation. However, individual pathways, such as Ras-ERK, are strongly related to cancer mutations and promise targeted therapies in the future[11].

The latest studies are now focusing on the tumor microenvironment as a critical element for determining tumor development, progression, and treatment response[5,8,12-13].

In the same research interest, artificial intelligence has multiple subsets or methods for data extraction and evaluation. One such method is artificial neural networking[5,12,14-16], which allows computational foundations, similar to neurons, to make connections and new neural pathways during data set training (See Figure 1). One such method used for quantitative biology is massive parallel

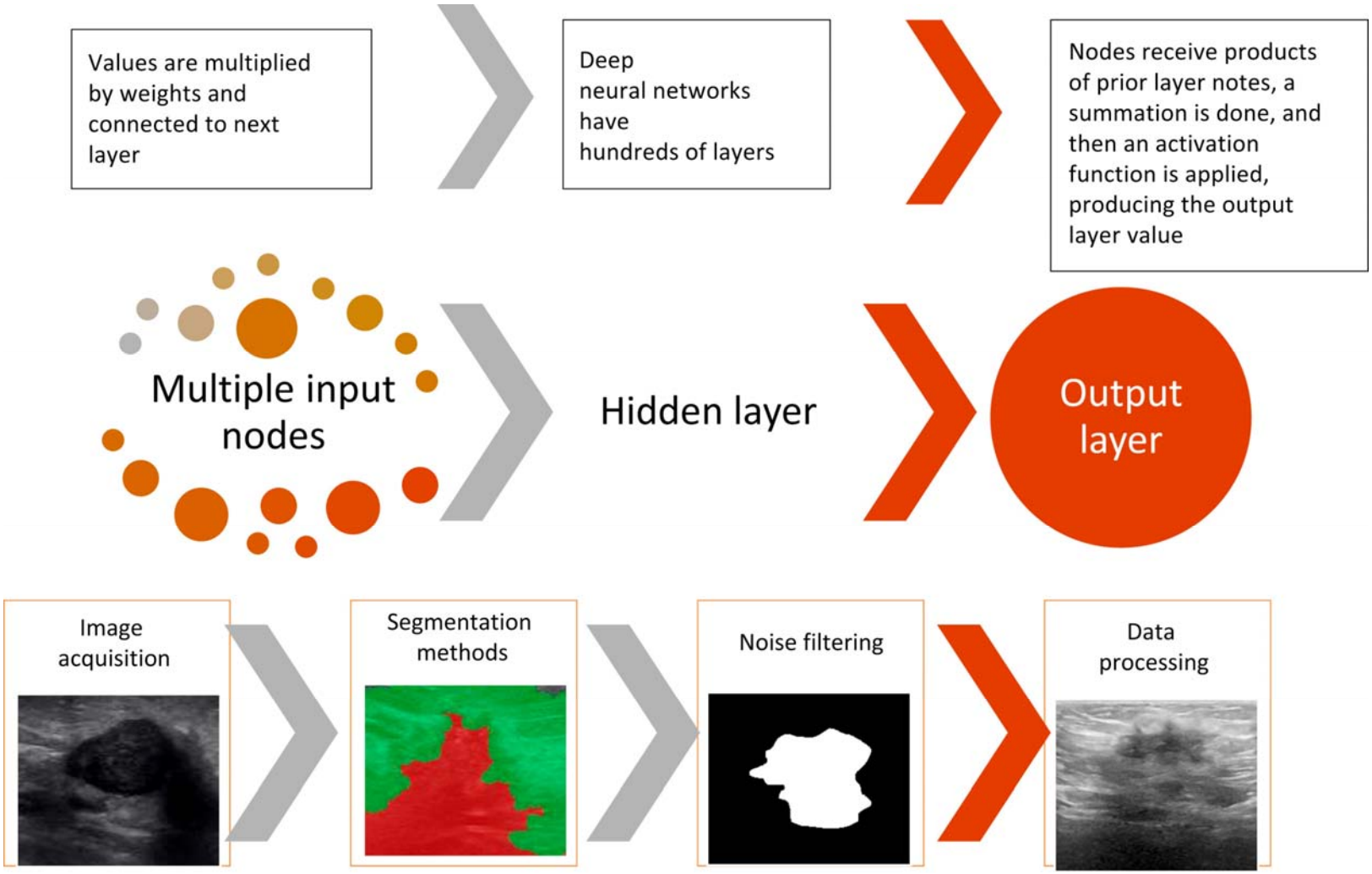

Figure 1. Illustration of mechanistic framework model.

reporter assay (MPRA), which assesses *DNA*[4]. This allows biologists the ability to predict molecular interactions and gene interactions. The mechanistic framework of gene regulation allows the possibility of new therapies to be developed[2,7-8,13]. There is a lack of congruence between biologists and Artificial Neural Networking (ANN) systems; the latest custom ANNs allow mathematical assumptions of common biological concepts so that the output is relayed as how a biologist would interpret results[6,12,14-16].

The first attempt at computerizing medical images[17] occurred during the 1960s, which is, to date, an important research topic in medical imaging with recent research delving into the artificial intelligence era for the medical field[18-20]. Computer-Aided Detection (CAD) serves as a diagnostic aid to support the physician's role by using non-invasive and accurate computer systems[17]. CAD incorporates quantitative analysis of images during the diagnostic process, proven from previous studies to increase the sensitivity of diagnosis by 21.2% and reduce the false-negative rate of diagnostic screening by 77%. Despite this figure, automated detection software is not widely used during breast screening[21]. A prospective study using CAD software during diagnosis has shown a 74% increase in cancer detection[21]. Certain technical advances in breast imaging—such as harmonic tissue imaging, compound imaging, and an extended field of view—have made its use integral during a breast cancer diagnosis. Standardized CAD techniques used in conjunction with ultrasound reduce the inter-observer variation.[20]

The detection rate of invasive cancers measuring less than 1 cm increases with the use of CAD systems. It can reduce false-negative rates from 31% to 19%,[22-23] in conjunction with dedicated breast imagers. The system assigns various sensitivity and specificity rates to cancers based on the lesion type. The sensitivity for malignant calcifications is 86–99% with CAD, with only 57% marked as amorphous calcifications[24]. The sensitivity for masses is estimated at 43–85%[25]. Further research is required to recognize suspicious asymmetries as they develop over time during serial imaging follow-up and assess the medico-legal implication of retained CAD-marked image information. A more extensive explanation of the various AI subtypes is discussed below.

Artificial Neural Networking (ANN) is the process of nonlinear mapping between set inputs to outputs. It achieves physical performance using dense processing elements similar to biological neurons. The ANN can learn and generalize from the examples given. Success is measured if complex linear functions govern the relationship between variables. Evolutionary computing consists of a collection of algorithms based on population evolution towards the solution of a problem. It is subdivided into genetic algorithms, genetic programming as well as evolutionary algorithms. Successful use is measured utilizing selecting features for classification of mammogram calcifications[4,10,26-28].

Overall the best approach is to combine these three main methods, for example, using a fuzzy logic system to design ANN evolutionary computing in automatic training and generating ANN architecture. Feature extraction can reduce an image to a small set of parameters called features (See Figure 4). The quality of a feature

depends on its contribution to detection, cancer classification, and the preprocessing steps and classification methods[17,27,29].

The quality of features cannot be categorized; due to the quality of a feature depending on its contribution to detection, classification, prognosis, and features dependent on its preprocessing steps and the classification measures. There are various types of features, such as geometric features, which refers to factors such as size and shape. The boundary is the starting point of extracting an object using AI. Various boundary methods are used, such as binary sets, which refers to the sets of pixels in a grayscale image, and edge detection, which defines an object by its edges. Other geometric features include area, volume, contrast, counting pixels inside an object boundary, perimeters as well as shape (no single shape descriptor can be used on its own to define an object)[15-16,28,30-32].

A computation method of predictive models through algorithms is referred to as machine learning. As more data is applied to the training data set, accuracy and predictability are optimized. Over the years, advances in algorithms and machine learning have allowed deep learning in recent studies. This has a similar output as the human brain's neural architecture, with neural nets responding to multiple data set training cycles using statistical frameworks. This learning method is ideal for image classification in radiology and pathology with above-average accuracy compared to human reader outputs[3,6,28,33-35].

The tumor microenvironment

The breast cancer microenvironment can be subdivided into three main subsets: local, regional, and distant. Each of these proposed subsets contains cellular contents such as fibroblasts, leukocytes, extracellular matrix, cytokines, growth factors, and hormones[7], described in detail below as the various cell type subsets related to breast cancer diagnosis and prognosis.

As Dvorak stated, tumors are much more than wounds that do not heal. Tumor cells undergo significant changes causing release from regulatory signals, promoting proliferation and invasion. The most crucial factor thereof is the overexpression of Vascular Endothelial Growth Factor (VEGF), allowing surrounding stroma to be incorporated in its progression process.[36]

The use of AI technology to improve diagnostic detection rates and remote disease monitoring can reduce the overall time required for overall patient treatment planning. Anti-VEGF agents and AI-generated prognosis have been studied using vision loss, which could promote the prevention of vision loss before its occurrence[37].

The angiogenesis process includes a complex interplay between tumor, endothelial, and stromal cells, promoting tumor growth. A study in 2006 found a novel method of assessing angiogenesis employing chick embryo and its chorioallantois membrane. An automated image analysis method was developed able to quantify the microvessel density and growth potential in images. This shows the potential to be used for tumor growth detection in breast cancer imaging,[6,8,14,36,38]; however, it lacks efficacy for extensive tumor series analysis of

TME. Other methods proposed for TME composition analysis are Gene Set Enrichment Analysis (GSEA- San Diego), xCell (California), and Timiner (Russia), which allows immunogenic analysis and quantification of the immune infiltrate[5,8-9,13,16,39-40].

GSEA[41] is a computational method able to define concordant differences between two biological states as a statistical output (See Figure 2)[41]. xCell[42] is a novel signature-based method used for 64 immune and stromal cell types. Utilizing in silico analyses and cross-comparison to cytometry immunophenotyping, xCell shows excellent promise when compared to other methods[42]. Timiner[43] is a computational pipeline used for the assessment of tumor-to-immune cell interactions based on sequencing data.

Anther computational method[44] in 2016 reported a Microenvironment Cell Populations counter (MCP counter) that analysis the transcriptomic markers in single-cell populations, but this method is robust compared to other used samples.[8]

The discussion below states the current body of knowledge and attributes of each factor/cell/protein related to breast cancer and the tumor microenvironment, followed by the latest technology and insights related to artificial intelligence and deep machine learning.

Figure 2. A method of gene expression signature profiling.

Fibroblasts and tumor progression

The vast majority of cells within the TME are fibroblasts, secrete various soluble factors modulating tumor stroma, growth, and invasion properties. Recent studies have found that cancer-associated fibroblasts have unique protein expression profiles making them unique in their identification properties. A bi-directional signaling pathway[9] has also been suggested between these unique fibroblasts and their adjacent cancer cells, suggesting a possible influence in the transcription of breast cancer cell profiles. This was also affirmed by Orimo et al[38], that cancer-associated fibroblasts enhance tumor angiogenesis.

These individual cells' origin has been suggested as either bone marrow, normal fibroblasts, and even epithelial-mesenchymal transition processes[7].

The microcellular environment is maintained by fibroblasts using remodeling of the extracellular matrix[9,36]. Fibroblasts, associated with carcinoma, have unique characteristics that promote tumor progression, presenting as either heterogeneous or myofibroblasts with fibroblast activation protein. The potential of carcinoma-associated fibroblasts promoting tumor growth uses secreted stromal-derived factor-1, acting as a paracrine activator that increases tumor cell proliferation through CXCR4[38].

An interesting finding was a co-culture of fibroblasts in healthy breast tissue "educating" fibroblasts to secrete HGF to promote tumor progression activities.

The main question that arises from these studies is: "where do these cancer-associated fibroblasts derive from?". One hypothesis is that healthy fibroblasts

undergo phenotype modification from constant aberrant signaling from adjacent tumor cells[45].

Dendritic cells (DC) and the role of Estrogen receptors

Dendritic cells play an essential role in prohibiting neoplastic cell growth by presenting antigens to CD4+ and CD8+ and T cells[7-10,13-14,36,38-39]. Dendritic cells' maturation process depends on their local microenvironment, which determines its tolerance of immunosuppression of localized neoplastic invasion. Surrounding tumor-associated stroma has shown in previous studies DC with an inability to stimulate antitumor immunity. These so-called tumor-associated DC produce proangiogenic factors, enhancing endothelial cell migration, causing tumor progression[6,8,14,36,38].

DCS has multiple roles in essential processes such as immunity, autoimmunity, and differentiation of T cells. They are mainly activated by stress response or pathogen-induced damage, which causes the secretion of cytokines stimulating T lymphocytes and immune response. Estrogen receptors play a crucial role in DC function[7-10]. When the DCS ligand binds to ERs, it triggers migration processes. Recent studies have shown that treatment of E2 alongside mature DCs and T cells could stimulate T cell proliferation[6,13].

Fibroblast, dendritic cells, and artificial intelligence

Fibroblasts play a role in mortality prediction of idiopathic pulmonary fibrosis (IPF). The use of AI to quantify prognostic histological features was studied, and found interstitial mononuclear inflammation and intra-alveolar macrophages proved as novel biomarkers in detecting IPF[46]. A group of researchers at Osaka

University developed an artificial intelligence-based system to identify various cancer cells utilizing microscopic images. A convolutional neural network was trained with 8000 images of cells obtained from a phased contrast microscope. Following the data set training process, another 2 000 images were tested to distinguish mouse cancer cells from human cells and radioresistant cells from radiosensitive ones. This study holds much promise in developing a universal system able to identify and distinguish between all variants of cancer cells[47].

Another researcher group based in Boston and Tufts Medical Centre, developed multiple artificial intelligence tools to detect and tract dendritic cells. An AI algorithm was developed using in vivo confocal microscopy (IVCM) analysis of the human cornea, typically done manually, making it a time-consuming process. The use of such AI models for analysis ensures high accuracy and reduced objectivity associated with human analysis[48].

Macrophages, lymphocytes and the role of Estrogen receptors

Macrophages associated with tumor cells display unique phenotyping, promoting tumor growth, angiogenesis, and tissue remodeling[4-5,7-10,12-16,36].

The immune response includes a key role of macrophages to promote T cell recruitment and activation. Their collaborative activation alongside T and B cells is due to cytokines, chemokines being released[7,14,39]. Despite their functional role in tumor defense, they are actively present in the tumor microenvironment leading to tumor progression and immunosuppression. Many articles report ER present in macrophage precursor cells during various stages of its differentiation process. E2 treatment has shown to change macrophage behavior[2-10,12,14-16,38-39].

A key player in recent research in the tumor microenvironment has been lymphocytes[16]. Lymphocytes are mostly T cells, CD4+ helper cells, T reg with CD4+, and CD 8+ cells.

T reg cells in the tumor microenvironment block its normal antitumor function and suppress other immune cells such as CD8+ T cells. T reg also produces a large amount of RANKL[4-10,12-16,33,36,38-39], which promotes metastasis and RANK-expressing neoplastic cells. A high concentration of T reg cells is associated with advanced type breast cancer. This is postulated as neoplastic cells recruiting T reg using prostaglandin E2 secretion, suppression effector cells, producing an immunosuppressive microenvironment.[4,7,9,15,36]

Macrophages, lymphocytes, and artificial intelligence

Machine learning (ML) can distinguish various cell and tissue types in a biopsy specimen based on a training set of "ground truth" examples. A research study in 2018 made use of ML algorithms as a method to identify macrophages from digital scans of non-small cell lung carcinoma tissue slides. The study compared pathologist output to the ML algorithm, which held improved accuracy compared to human- reader intervention and output.[49]

A Working Group collaboration with the Massive Analysis and Quality Control Consortium works on machine learning algorithms to characterize tumor-infiltrating lymphocytes. Such methods will enhance the validity of prognostic prediction methods in pathology. besides the clinically evident improved prediction rates of ML, it also permits changes to the current feature set used for

ML analysis, thus improving accuracy and interpretation to current standard methods.[50]

Extracellular matrix (ECM), Mast cells, and neutrophils

The main proteins within the complex ECM are collagen (structural), fibronectin (glycoproteins), and chondroitin sulfate (proteoglycans). Recent studies have shown that ECM[9] is more versatile than initially thought, acting as a critical player in cell growth, proliferation, and migration. In cancer, ECM is typically disorganized in appearance, causing abnormal feedback regulatory mechanisms. This is mainly due to ECM metabolism being altered by CAF and immune cells[5,8-9,13,16,39]. One of the main proteins within ECM, namely collage, promotes cancer cell invasion using collagen IV degradation. ECM also promotes the passage of cytokines and growth factors, enabling intercellular communication. The alteration in protein activity is seen in cancer is associated with patient outcomes.

Mast cells (MCs) form part of the immune system, associated with parasitic infections. Depending on the type of inflammatory stimulus, mast cells release various inflammatory mediators. Mucosal MCs produce tryptase, whereas connective tissue MCs secretes tryptase, chymase, and carboxypeptidases. All of these enzymes, along with IL-8, TGF-β, and TNF-α, have a strong association with angiogenesis and MMP modulation of various breast cancer phenotypes.[5,14-15]

ERα has been found evident in mast cells. The treatment of E2 has shown, in rat mast cell models, a release of histamine. This process is enjoyable because histamine release plays a role in breast cancer promotion utilizing its H3R and H4R receptors. [6,13]

Neutrophils are a fundamental component of the immune response, acting as a first-line defense mechanism against infection employing phagocytosis. Neutrophils work alongside other immune-fighting cells such as macrophages and Dendritic cells (DCs).[5,8-9,13,16,39]

Neutrophils are known to have nuclear receptions, and E2 and ER binding help regulate neutrophil survival and function. Several serine proteases are secreted by neutrophils, such as neutrophil elastase (NE), proteinase 3 (PR3), and cathepsin G (CG), essential for infectious agent elimination and inflammation modulation.[7,36]

Mast cells, extracellular matrix, neutrophils, and artificial intelligence

One of the critical elements of mast cell granules is histamine, as it has been shown to promote tumor cell proliferation and growth of mammary carcinomas through H2 receptors[51]. A study attempting to assess through machine learning functional genomic networks discovered histamine hypersensitivity in response to a local inflammatory response, which begs the question of its underlying molecular and genetic traits and how machine learning could promote its prognostic indicators role in tumor progression[52]. Many research studies coin mast cells as the most misunderstood cell type during breast cancer proliferation and immune response since its discovery 140 years ago, making them a key focus of future research endeavors[53].

Advances in 3D cell tissue engineering have led to the development of "cancer on a chip" platforms, which allow the TME model to have improved analytic outputs, especially for discovering the role of the extracellular matrix during tumor

progression. The possibility of integrating artificial intelligence for improved drug screening models is made possible through these chip platforms[54-55].

Microscopy has reached the age of digitization with outputs such as CellaVision, which classifies degenerated lymphocytes and web-like remnants. The researchers hypothesize that these remnants are Neutrophil Extracellular Traps (NETs). They aim to develop an AI platform able to detect NETs rapidly on blood smears[56].

The use of computational models to screen endocrine-active compounds holds much promise as a cost-effective alternate method in practice. A machine-learning algorithm was applied to over 7500 compounds related to nuclear estrogen receptor (ERα and ERβ) activity. The model's performance was evaluated using receiver-operating curve values obtained from fivefold cross-validation procedures, which proved values ranging from 0,56-0,86[6,35].

The following sections elaborate on the surrounding environments related to breast cancer and TME. Each section will discuss the current trends and research and the latest AI technology being developed.

Breast cancer and local microenvironment

Normal mammary gland development relies on appropriate cross-talk between epithelial and stromal cells, inhibiting abnormal cell growth and neoplasm formation. Myoepithelial cells have previously been known for their tumor suppression capabilities as they produce a base membrane barrier around luminal epithelial cells. The loss of such myoepithelial cells would promote in-situ carcinoma to invasive type carcinoma[4,7-8,10,38]. Two models have been suggested to explain this carcinoma invasion. The 'escape model' suggests genetic changes of tumor epithelial cells, allowing the invasion to adjacent ducts. The 'release' model suggests that the tumor microenvironment disrupts the basement membrane, allowing tumor cells to spread into the stroma (See Figure 3). Figure 3 describes the concept of escaped immune cells during the transition from in situ to invasive carcinoma. Both of these models prove the importance of both epithelial and stromal components in tumor progression.[39]

Artificial intelligence and the local microenvironment

In the last decade, many approaches have been used to quantify the non-cancerous cell populations from acquired tumor samples, using computational algorithms with different statistical frameworks and data sets. The two most common algorithms used for TME estimation are regression-based deconvolution algorithms and gene-set enrichment methods. The algorithms are dependent on pre-acquired knowledge of the data sets for accurate measurement, a statistical framework, and a pre-determined signature for each cell type. The regression-based deconvolution algorithm determines the gene expression profile ratio in

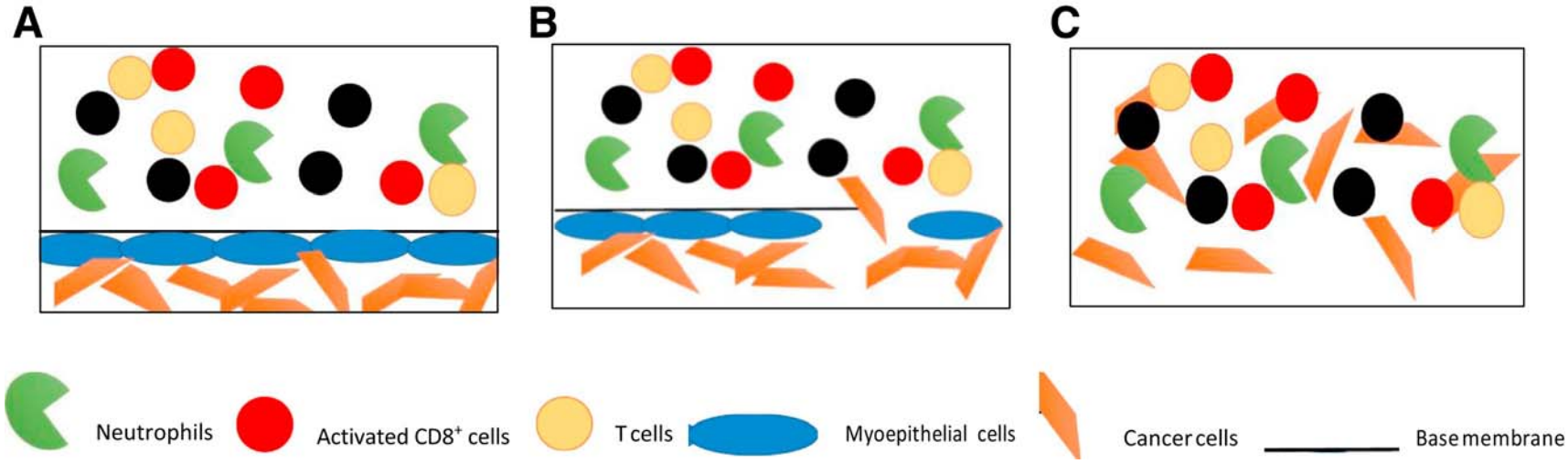

Figure 3. Illustration of escape model from *in situ* to invasive carcinoma. **A:** *In situ* carcinoma and the immune environment. **B:** Locally invasive *in situ* carcinoma. **C:** Invasive carcinoma causes enrichment of TREG gene sets and less activated $CD8^+$ T cells.

the total tumor expression profile. Gene-set enrichment assigns scores to the various cell types as a function of its expression in each gene set[57].

A recent study of the University of Eastern Finland[6] developed an AI model capable of predicting breast cancer risk based on demographic risk factors and genetic variants. The method used for the AI model is a gradient tree with adaptive iterative searching methods. The gene interaction map included ESR1 an FGFR1 genes[2,4-9,13-16,36,38,58], linked to estrogen receptor subtype breast cancer. Since cancer incidence is a multifactorial process, the use of AI in predicting breast cancer risk through this novel method holds much promise for future disease incidence[6,12,15].

Breast Cancer and the Metastatic microenvironment

During the complex metastasis process, tumor cells either have a dormant state or an active state of forming micrometastases. During the primary tumor recruitment, the cytokines select associated bone-marrow cells to incur a premetastatic process before tumor mobilization occurs. It has been noted from previous studies that fibroblasts and cancer cells travel alongside one another during the metastatic process. Breast cancer cells promote receptor activate nuclear factor κβ ligand (RANKL) through active secretion of cytokines and growth factors[4,6-7,9,13,36,38]. This activates osteoclast formation and bone resorption. RANKL has more recently been noted in the formation of lung metastases, thus providing a hypothesis of specific immune cells partaking in metastases' formation.

The seed planted in the soil concept confirms that malignant disease remains the foundation of a tumor progressing, whereas the tumor microenvironment facilitates these cancer cells' invasion ability. For this reason, research currently focuses on epithelial-mesenchymal transition (EMT), where specific mediators allow the progression of tumor cells to invasive type lesions. Examples of these mediators are IL-1, IL-6, and IL-8, which allow tumor cell proliferation with EMT increasing their ability to metastasize[4,7,16,38].

A fundamental attribution to tumor progression and drug resistance is the tumor microenvironment. This implies that various ill-controlled cells all relate to cancer progression. This concept has been around since the 1880s, where Steven Paget suggested the "seed and soil" concept[59], where fertile soil (the tumor

microenvironment) and the seed (cancer cells) work in harmony to promote growth.

Both intrinsic and extrinsic inflammatory pathways promote an inflammatory microenvironment. Tumor cells promote inflammatory mediators, which leads to the progression of cancer within the microenvironment using T cells, NK cells, macrophages, and dendritic cells.[36]

Artificial intelligence and the metastatic microenvironment

Recent AI insights allow assessing molecular subtypes and their therapeutic response utilizing predictive image analysis of breast cancer phenotypes.

In a research study of the TCGA Breast Phenotype Group[5,15], multidisciplinary researchers phenotypically characterized 84 solid breast tumors to gain insights into the underlying molecular characteristics and gene expression profiles. Significant similarities were noted between enhancement texture (entropy) and molecular subtypes (normal-like, luminal A, luminal B, HER2-enriched, basal-like) even after controlling for tumor size ($P = .04$ for lesions ≤2 cm; $P = .02$ for lesions from 42 to ≤5 cm). [14-15,36,39,59]

Regarding treatment outcomes, a semi-manual delineation method of tumor volume using breast MRI imaging proved a high prediction anomaly for low recurrence rate in patients, proving the potential for digital automation in its prediction outcomes.

Breast Cancer and infiltrated immune cell microenvironment

Significant gene expression changes occur within myoepithelial cells, confirming a change during tumor progression in the microenvironment. An example of overexpression of genes is chemokine CXCL14, binding to CXCR4, promoting proliferation and migration of tumor cells. Others also confirmed this study that changes in the stroma and gene expression occur most frequently when healthy breast tissue transitions to DCIS. [7,38,58]

Since breast cancer is a heterogeneous disease, it has three main phenotypes: luminal, human epidermal growth factor receptor type, and triple-negative type. Since breast cancer promotes an inflammatory microenvironment, immune filtration is presently based on Estrogen Receptor (ER) presence[7,13,33,38]. There is a substantial proportion of natural killer cells and neutrophils within ER-positive breast cancer and cytotoxic and TCD4+ cells in smaller amounts. The presence of eosinophils, monocytes, and B lymphocytes proved a good prognosis following chemotherapy.[5,8,39,59]

Artificial intelligence and molecular alterations of the microenvironment

Despite the significant potential AI holds, there is still much debate surrounding the appropriate and ethical curation of medical data from Picture Archiving and Communication Systems (PACS). The clinical significance of AI output still holds its outcome based on its human predecessor's data training sets.

The integration between biomarkers, risk factors, and imaging data will allow the best predictor models for patient-based outcomes.[5,7,60]

State of the art research has found a machine learning approach, named CytoReason(Version 1.0)[1], distinguishing between nivolumab responders and non-responders. Since adipocytes are postulated to be involved in the tumor microenvironment, this study also showed evidence of their regulatory role in ipilimumab resistant nivolumab patients. The study requires extensive research on adipocytes' role in tumor progression, leading to new immunotherapy methods. Cytoreason[1] integrates genetics, proteomics, cytometry, and literature with machine learning to help create disease models.

A key focus on T-cell subsets related to cancer immunology and therapy is adamant as a prediction of such subsets could promote advances in immunology research. A research group developed a method, Immune Cell Abundance Identifier (ImmuCellAI – China), which allows gene set signature-based algorithms to estimate the abundance of 24 immune cell types from gene expression data[61]. However, the method has limitations, such as measuring the abundance of cells being limited to the deviation from gene signatures. The method also did not include spatiotemporal attributes of the immune cells[61].

A wild-type adeno-associated (AAV) particle capsid is currently the most commonly used gene therapy method due to its established ability to deliver gene material to organs. However, few naturally derived AAV capsids are deficient in the essential components required for gene therapy. A group of researchers at Harvard developed machine learning technology to engineer new, improved capsids for therapeutic use.

Starting at Harvard in 2015, the authors set out to overcome the limitations of current capsids by developing new machine-guided technologies to rapidly and systematically engineer a suite of new, improved capsids for widespread therapeutic use, which outperformed AAVs generated by conventional random mutagenesis approaches. This demonstrates a powerful tool for sizeable broad-scale *DNA* synthesis, iterative machine-guided design to develop improved synthetic AAV capsids[62-63].

The current and latest trends to local, metastatic, and microenvironment hold much potential for the forthcoming years in breast cancer research and AI technology. The next section discusses the most recent research and trials, which have been done in the past year of 2020.

Artificial intelligence and beyond

A recent 2020 study in Italy[16,34] focused their research on predicting the disease, establishing a therapeutic plan, and patient-focused follow-up sessions. In this regard, a multidisciplinary approach has been encouraged during the development of the Multigene Signature Panels and Nottingham Prognostic Index. Machine learning allows the cross-correlation of prognostic indicators to determine possible markers related to patient outcomes. Two machine learning methods were deployed, namely Artificial Neural Networking and Support Vector Systems using SPSS IBM Modeller 18.1 software. Their accuracy, sensitivity and specificity was measured as 95,29%-96-86%, 0,35-0,64 and 0,97-0,99 respectively[34]. The study was limited to a select study population without long-term recurrence following 20 years of remission in breast cancer patients.

Breast cancer comprises a complex genetic background, and the intricate relationship between these cancer cells and surrounding stromal/immune cells is essential to ensure adequate treatment methods are implemented. In vitro cell culture systems lack the dedicated physiological outputs during drug testing[8,38]. Mouse models have the ideal animal model for assessing drug tolerance; however, it is limited in testing humans' tumor microenvironment. Various models have been proposed for tumor microenvironment studies, where the latest in vitro 3D models can study both cell-cell and cell-material interactions parametrically.[8,38]

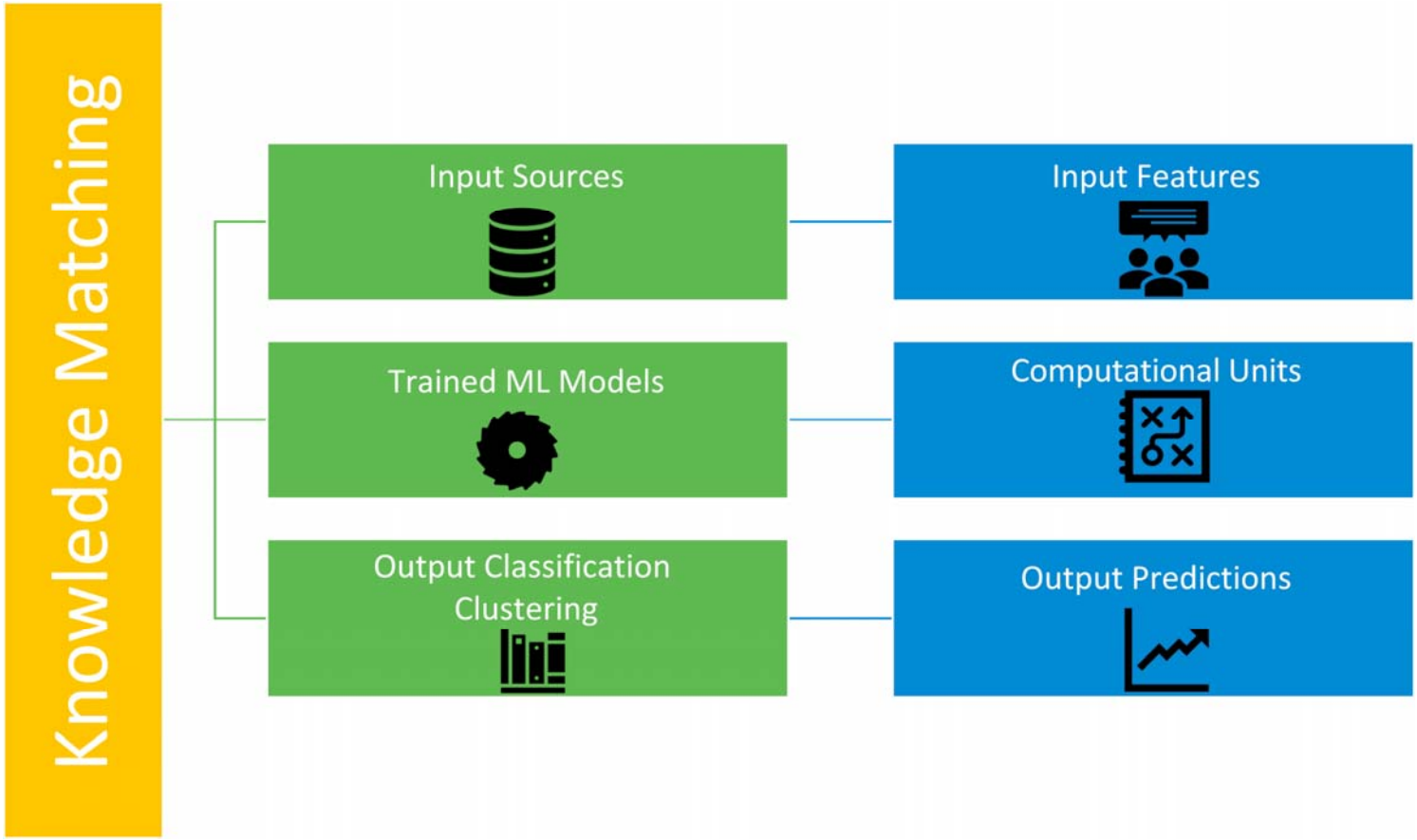

Figure 4. Illustration of ML mechanistic framework. ML, machine learning.

The use of stromal-to-epithelial yield using spatial extraction of features is also a novel approach to assessing disease progression. These studies allow further insights into the role of epithelial and stromal cells and an alternate tiered approach to deep machine learning.[4,9-10,14,16,36,38-40,44,59]

The main answer all therapists, pathologists, and clinicians require is an improved prognosis method in breast cancer. Shimizu and Nakayama[64], developed a complete atlas of prognostic breast cancer genes, a computational framework and prediction score, applicable to all breast cancer subsets. The method is unique in its stratification of patients at the clinical stage and estrogen receptor-negative subtypes[64].

Conclusion

The use of tissue engineering[65] in cancer research allows an accurate representation of TME in human studies[5,10,15]. Since there is currently vast recognition of TME in tumor progression, it is now the current therapeutic research focus. New strategies to normalize the surrounding stroma, modulation of the immune system, and antitumor activity enhancement are evident. The critical role of E2 and its signaling pathways requires more research on the use of intertumoral therapy as part of an adjuvant therapy approach to immune response.

Despite some limitations in mouse models, the data supports the role of TME in the treatment of breast cancer.[4,6-7,9-10,13,15,33,66]

The various tumor microenvironment elements and their latest research endeavors using AI and DML shows that the purpose of improved prognostic and therapeutic methods is adamant. The role of unique cancer signaling pathways, targeted therapies, and novel diagnostic trends will boast significant strides when conjoined with the AI, as mentioned earlier in practice. The development of a comprehensive prognostic cancer gene mutation atlas will be a step into the future generation for pathologists, even more so as a multidisciplinary approach for developing the Multigene Signature Panels and Nottingham Prognostic Index. Although most studies are experimental and within clinical trials, the possibility of integrating such methods in clinical practice is an almost certain future.

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7.) CORRESPONDING AUTHOR'S DETAILS

Kathryn Malherbe

Kathryn.malherbe@up.ac.za,

Department Radiography, Faculty Health Sciences, University of Pretoria.

+27716732188