cancers

Review

Machine Learning for Future Subtyping of the Tumor Microenvironment of Gastro-Esophageal Adenocarcinomas

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Simple Summary: We summarize the main components of the tumor microenvironment in gastro-esophageal adenocarcinomas (GEA). In addition, we highlight past and present applications of machine learning in GEA to propose ways to facilitate its clinical use in the future.

Abstract: Tumor progression involves an intricate interplay between malignant cells and their surrounding tumor microenvironment (TME) at specific sites. The TME is dynamic and is composed of stromal, parenchymal, and immune cells, which mediate cancer progression and therapy resistance. Evidence from preclinical and clinical studies revealed that TME targeting and reprogramming can be a promising approach to achieve anti-tumor effects in several cancers, including in GEA. Thus, it is of great interest to use modern technology to understand the relevant components of programming the TME. Here, we discuss the approach of machine learning, which recently gained increasing interest recently because of its ability to measure tumor parameters at the cellular level, reveal global features of relevance, and generate prognostic models. In this review, we discuss the relevant stromal composition of the TME in GEAs and discuss how they could be integrated. We also review the current progress in the application of machine learning in different medical disciplines that are relevant for the management and study of GEA.

Keywords: gastric cancer; esophageal cancer; gastro-esophageal; machine learning; tumor microenvironment; deep learning; artificial intelligence; immunotherapy; omics

1. Gastro-Esophageal Adenocarcinoma (GEA)—An Introduction

1.1. Tumor Microenvironment (TME)

Tumors may be seen as an abnormal organ, forming as a result of close interaction of cancer cells with the surrounding tissue [1]. Cancer initiation is a process primarily driven by genetic alterations of somatic cells at the site of tumor origin, but concomitant responses occur at the cellular level altering the TME [2,3]. For instance, increased expression of pro-inflammatory cytokines such as interleukin (IL)-1, IL-6, interferon (IFN)-γ, and tumor necrosis factor (TNF)-α, leads to recruitment and activation of several cell types of immune and stromal cells that promote adaption of residual cells [4–6]. As tumors grow, genetic alterations may increase in complexity [7]. In parallel, imbalances in nutrient supply (hypoxia), as well as acute-to-chronic inflammation reveal a dynamic shaping of this process [8,9]. Therefore, using snapshot information on the TME appears unlikely to be useful, as the TME is dynamically altered by multiple factors (Figure 1). These complex factors include nutrient supply, genetic alterations, and cytokine/chemokine gradients, all of which can show temporal and spatial intratumoral heterogeneity [10,11]. As current cancer treatment paradigms are shifting from "one-size-fits-all" therapeutic strategies to approaches

based on precision medicine, it is of crucial importance to gain a relevant understanding of the TME to suggest the best therapy for every single patient at an individual level and specific stage of progression, as well as to discover novel therapies in the near future.
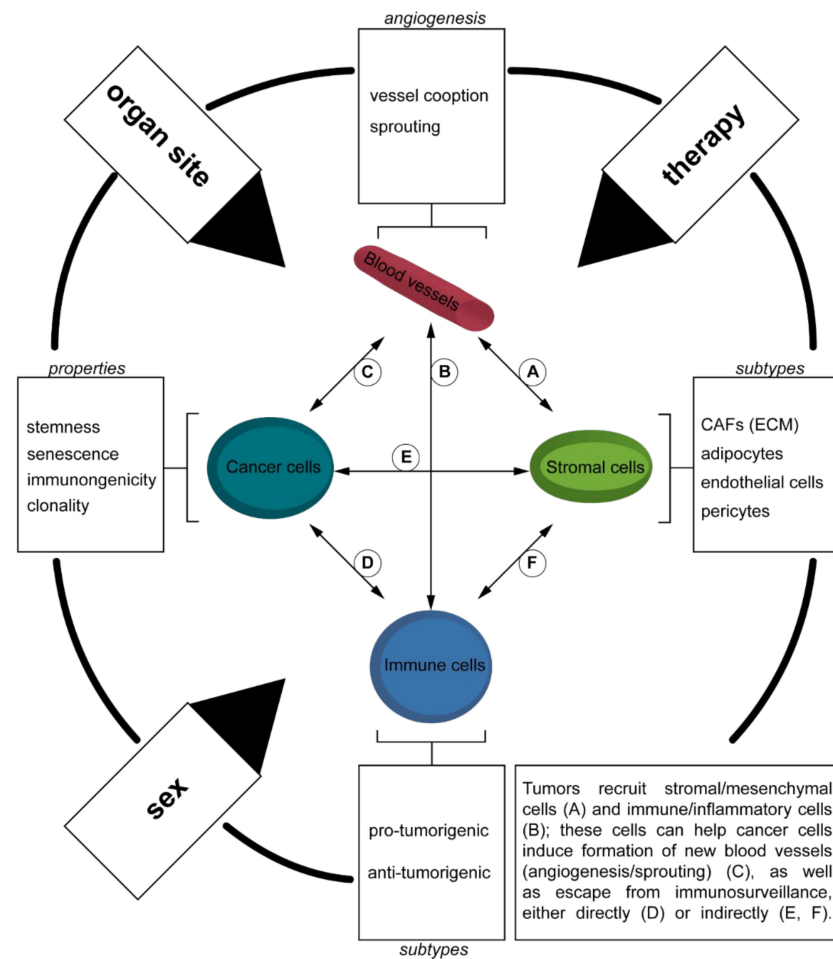
Figure 1. Integrative view of the tumor microenvironment. During cancer development, somatic mutations are acquired at the DNA level leading to uncontrolled cell growth. In detail, tumors are formed with clonal heterogeneity and potential stem-cell-like properties, in line with immune cell exclusive properties [9,12,13]. Within the context of the regional parenchyma (site of origin; localization), cytokines are released and promote new vessel formation, a process that may involve both sprouting angiogenesis as well as the co-option of preexisting vasculature, among other mechanisms [14–16]. The newly developed vasculature is usually immature and presents abnormalities, including increased permeability and poor perfusion (due to lack of pericyte coverage or due to collapse because of surrounding physical stress, which is deposited by specialized cancer-associated fibroblasts or CAFs) [6,17]. Vascular function is also influenced by the excessive deposition of extracellular matrix (ECM) components which may lead to blood vessel compression, altering oxygen supply, and decreasing therapeutic delivery and efficiency [18]. Hypoxia may increase genomic stress in cancer cells, in addition to other characteristics of cancer progression and therapy resistance [7,19]. In addition, the abnormal characteristics of blood vessels will attract inflammatory cells [20,21]. Moreover, cytokines and chemokine expressed by the cancer cells may attract immune cells, including lymphocytes, granulocytes, and macrophages, shaping a pro-tumorigenic TME, a process that may be influenced by sex in GEA [22–27] and administration of (cytotoxic) therapies [28,29], underlining a connective network between tumor cells, stromal cells, immune cells, and blood vessels. Together, as tumor site may change (as the progressing tumors metastasize at distant sites) and therapies increase selection pressure, the TME may undergo dynamic changes [30]. Moreover, administration of (cytotoxic) therapies potentially selects for cancer cell traits leading to senescence, a potential mediator of disease relapse [31].

The TME of established solid tumors consists of different cellular components, including activated fibroblasts [32,33], immune cells (lymphocytes, macrophages, dendritic cells), and endothelial cells, all with distinct functions, reviewed elsewhere [34–37]. Other mechanisms in early tumorigenesis and late disease progression include epithelial-to-mesenchymal transition (or EMT), a process whereby tumor cells may undergo specific phenotypical changes, which is reviewed elsewhere [12,13].

The therapeutic promise brought by new immunotherapies, in particular those targeting immune checkpoint molecules or administration of antitumoral immune cells, has shifted the interest of TME studies towards a better understanding of the immune TME [13]. Here, the abovementioned cellular components can show distinct phenotypes and anti- or pro-tumor activities, in a cancer type- and site-dependent manner [14].

We will briefly discuss methodical approaches using machine learning (ML) that may allow to provide a more integrative view on the TME and facilitate this deeper understanding. We performed a focused publication search using PubMed database by using keywords such as tumor microenvironment, gastric cancer, esophageal cancer in addition to a dedicated search for literature on the topic of machine learning using the following search query: (gastric cancer OR esophageal cancer OR gastro-esophageal cancer OR gastro-esophageal junction cancer OR esophageal adenocarcinoma) AND (machine learning OR artificial intelligence OR deep learning).

1.2. Brief Overview of GEA

1.2.1. Introduction

Worldwide, the incidence of gastric cancer ranks 6th and the number of related deaths ranks 3rd; esophageal cancer ranks 10th and 6th, respectively, according to the global cancer statistics of 2020 [15]. Pathologically, adenocarcinomas of the lower esophageal tract and gastric adenocarcinomas can have diverse etiologies. Adenocarcinomas of the esophagus are primarily thought to develop as a result of gastro-esophageal reflux disease [16–18], while adenocarcinomas of the stomach may arise in association to infection with Epstein–Barr virus or helicobacter pylori or may develop in a setting of cancer predisposition syndrome—which is discussed elsewhere [17,19,20]. However, there is often a mutational overlap between gastric and esophageal adenocarcinomas [21]. Molecular classifications include esophageal adenocarcinomas into the chromosomally unstable subtype of GEAs [22,23]. Clinically, the standard therapy for GEA includes perioperative therapy, surgery, and radio-chemotherapy depending on tumor stage and patient characteristics with additional potential novel therapies which are discussed in the following section [24].

1.2.2. Current and Future Therapeutic Concepts in GEA

Despite targeting oncogenic alterations of tumor cells in GEA, progress has been limited in unselected patient populations [21,23,25]. Targeting the TME has become of great clinical interest, fueled by the development of effective anti-angiogenic therapies and immunotherapies [26–31].

Although initial trials with the anti-vascular endothelial growth factor (VEGF) antibody bevacizumab initially provided negative results, the anti-VEGF receptor (VEGFR)-2 blocking antibody ramucirumab demonstrated efficacy in GEA alone and with chemotherapy [38–40]. Interestingly, in a recent meta-analysis, anti-angiogenic therapies added a benefit to overall survival in these cancers. It is also worth noting the recent positive results with the use of the multitargeted tyrosine kinase inhibitor (mTKI) apatinib, an agent with anti-VEGFR activity, in Chinese GEA patients [41,42].

More recent efforts in oncology have centered on the development of therapies that use immune-checkpoint inhibitors (ICI) [26,28,43]. A key mechanism of action of ICIs is to alleviate immune-cell exhaustion that leads to immunogenic tolerance towards tumor cells. Blocking these molecules would enhance anti-tumor immunity, and indeed, this concept is supported by recent successes in GEA patients [44–47]. This has led to the approval of ICI in advanced stage GEA cancers expressing PD-L1 [48–50]

A particularly promising therapeutic avenue appears to be the combination of ICIs with anti-angiogenic agents via normalization of the vasculature and reprogramming of the immune TME, reviewed elsewhere [51–56]. Interestingly, this concept is now being evaluated in several clinical trials in GEA with promising initial results [51,57].

1.2.3. The TME of GEA

Chronic inflammation can be seen as a major risk factor for developing GEA [58–61]. Here, cytokines released both locally and systemically (for example in patients with underlying conditions such as obesity) create a disbalance of cellular stress [62–65]. In the process of tumor initiation, several cytokines shape a pro-tumorigenic TME with accumulation of myeloid-derived suppressor cells (MDSC) in the very initial phase, in addition to macrophages, reviewed elsewhere [36,66–71]. For instance, the pro-fibrotic and immunosuppressive transforming growth factor (TGF)-β, or the pleiotropic immune mediators IL-1 and IL-6, mediate this process [5,6,72–75].

With more men than women being affected by GEA (6.6 [8.2]/1.8 [3.8], region-specific incidence for age-standardized rates by sex for esophageal cancer [stomach] in 2020, Western Europe [15]), one should also appreciate that sex may be considered as a variable in future trials and clinical management of GEA patients (Figure 1) [76–80]. Given the relevance of sex differences in cancer mortality, molecular and genetically, as well as pharmaceutically, future studies need to define the underlying mechanisms for these differences when studying the TME of GEA [81].

1.2.4. Biomarkers in GEA

As precision oncology emerges, classification of GEA such as the TCGA—chromosomal instable subtype (50%), microsatellite instable (MSI) high subtype (22%), genomically stable subtype (20%), Epstein–Barr virus-positive subtype (9%) among other classifications—are increasingly being used for prognostication [21,82–86]. Unfortunately, these classifications have not yet been fully translated into improved therapeutic regimens, although MSI high subtypes and Epstein–Barr virus-positive cases show increased rates of response to ICI [21,82–86]. Additional genes that may help GEA patient stratification for targeted therapies include *EGFR* and Her2/neu (*ERBB2*) [87–93]. Finally, tissue biopsies, taken at initial diagnosis, may also be used to identify expression signatures to predict response primarily to neoadjuvant therapy [94–98].

Given the increasing role of immunotherapy in the treatment of GEA, inflammatory phenotypes and biomarkers that are linked to pro/anti-tumoral properties are investigated in current studies [99,100]. Mechanistically, acquired, or intrinsic resistance to immunotherapy is a complex process. Thus, a generic biomarker to predict response to ICI remains elusive. However, PD-L1 combined positive scoring (CPS), appears to identify patients with GEA that may respond to anti-PD/L1 antibody immunotherapy. In addition, there is interest in defining the role of the number of somatic mutations (tumor mutational burden) as a biomarker for ICI [48,101,102]. Many prediction models of response to ICI consider the frequency of tumor-infiltrating lymphocytes (TILs) [4,103–106]. Moreover, integrative diagnostic approaches that combine several omics techniques have been shown to increase prediction to response to ICI therapy, including tumor-mutational burden (TMB) or neoantigen burden, to identify tumors with pro-immunogenic properties [102–104,107–111].

2. Machine Learning—Basic Concepts, Specific Applications, and Future Directions in GEA

2.1. Basic Concepts of ML

ML has gained recent interest within medical research, as large (annotated) datasets have become available, hardware components have allowed more complex models to be trained and a broad distribution and accessibility of code and examples have emerged and allowed the field to grow rapidly. Within the following section, we will first introduce the basic concepts of ML and then briefly review their application in GEA.
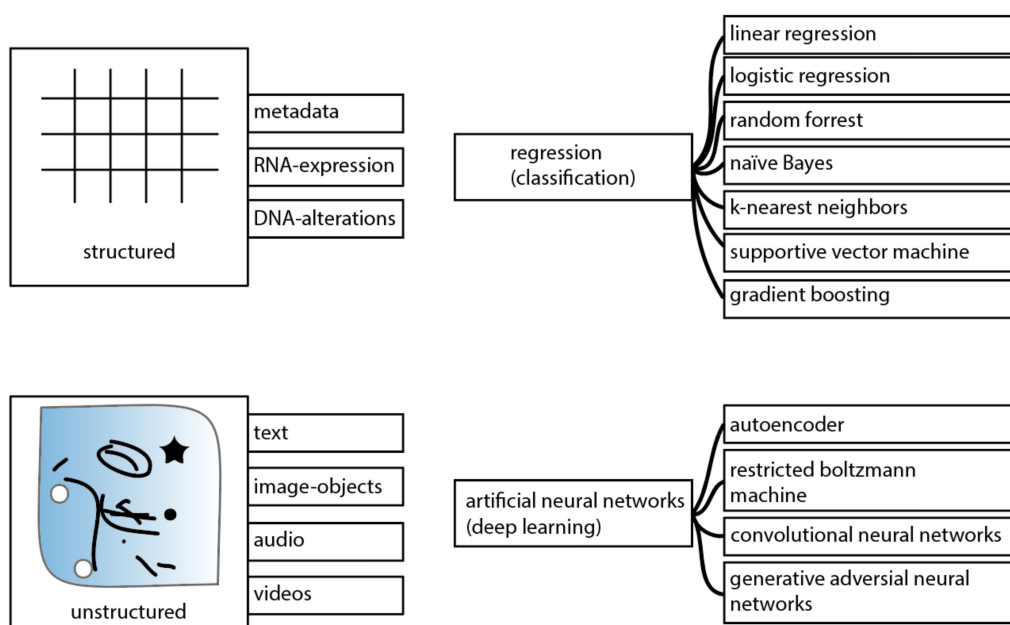
2.1.1. Supervised Learning

The term "supervised" refers to the technique where a model is supplied with data (known as features; for instance, genes with quantile normalized array data) and a target variable is defined (outcome; for instance, response to therapy). Depending on the design of the algorithm and the nature of the type of the target, the algorithm may return a class (responder/non-responder) or a continuous variable (time to relapse/score). The application of supervised learning with certain deep convolutional neural networks is occasionally referred to as artificial intelligence [112].

2.1.2. Unsupervised Learning

Here, no target variables are defined that a given algorithm is trained on. Rather than proposing classes or scores, unsupervised learning methods are primarily used to show (visualize) differences and similarities between samples. Commonly, unsupervised learning is used to reduce the complexity of a dataset for subsequent supervised learning (feature selection). However, within data exploration, unsupervised learning may be applied to study relationships and understand connections that need to be uncovered in each dataset, including gene network analyses [113,114]. For instance, visualization techniques using principal component analysis (PCA) and *t*-statistic stochastic neighbor embedding (t-SNE) are widely applied in the biomedical field, especially given the growing interest in single-cell RNA/DNA sequencing [115–118].

2.1.3. Choosing the Right Approach for the Right Kind of Datatype

The application of different DL models for supervised learning has allowed major advances to within the biomedical field (Figure 2). Especially for object detection, classification, and (semantic) image segmentation DL allowed major progress to be made. Although DL shows advantages to solve problems of unstructured data, classical regression and classification models are still useful. Linear regression models have the advantage to allow revealing the contribution of variables. This can be of interest (in the medical field) to potentially allow quality control of the variables of interest or to even collect the given variables actively for future studies.

Figure 2. Overview of ML techniques that can be applied using an unsupervised learning approach. Regularly, tabular data (structured data), including genomics data are analyzed using regression or classification models. Notably, also structured data can be analyzed using deep learning (DL). As for unstructured data, where complexity increases, DL models are used in favor of regression/classification models. In particular, the field of computer vision and image analysis has shifted greatly to DL.

2.2. Specific Application of ML in GEA

So far, several studies have applied regression models and DL models to address different kinds of medical problems in GEA. We have divided the different disciplines and diagnostic modalities to show the potential application of ML for GEA. A summary of the disciplines and the type of problems addressed by them are summarized in Figure 3.
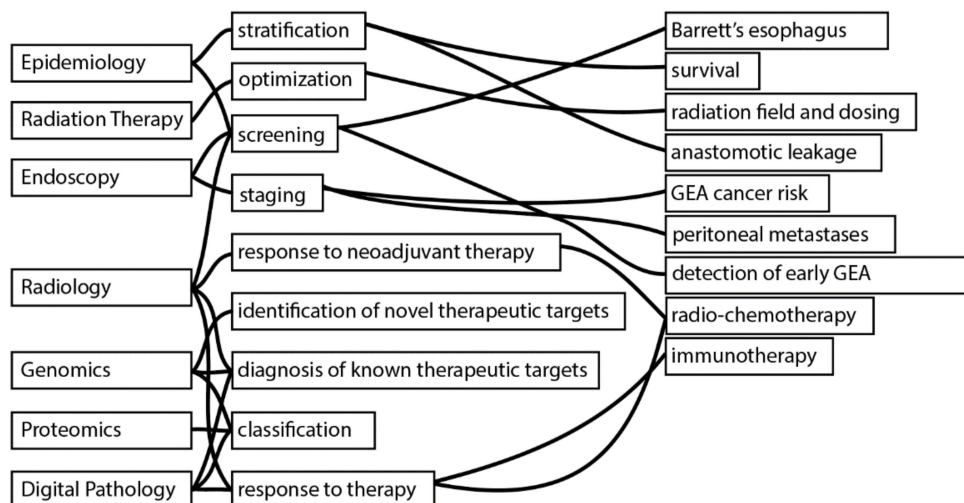
Figure 3. ML applications according to medical disciplines and diagnostic modalities in GEA. A description of studies following the given examples can be found in Sections 2.2.1–2.2.5. In summary, medical disciplines and diagnostic modalities including Epidemiology, Radiation Oncology (Therapy), Endoscopy, Radiology, Genomics, Proteomics, and Digital Pathology have shown how ML can be used to stratify patients for survival and complications of surgical intervention; optimization for dosing and radiation fields; screening for Barrett's esophagus (dysplastic/non-dysplastic); early GEA detection; staging of cancer (peritoneal metastases, lymph node metastases); response to (neoadjuvant) therapy (radio-chemotherapy/immunotherapy) and discovery/diagnosis of novel/current therapeutic targets.

2.2.1. Epidemiology, Radiation Oncology, and Blood Biomarkers

Yoon et. al., used a logistic regression model and a supportive vector machine to predict excessive muscle loss during neoadjuvant radio-chemotherapy by analyzing patients' blood samples and body mass index [119]. Interestingly, ML may also be used to propose risk factors for anastomotic leakage after esophagectomy [120]. Other attempts included using DL to identify optimal dosing of radiotherapy in GEA or defining the optimal target volume and organs at risk [121–125]. A dedicated analysis by Rahman et. al., used a random survival forest model by utilizing a dataset of more than 6000 patients to identify long-term survivors after esophagectomy [126]. Aslam et al., applied an autoencoder to a breath analysis and showed that this approach may be used for early GEA detection [127].

By applying an Extreme Gradient Boosting (XGBoost) technique, Leung et al., predicted the risk of GEA development after *Helicobacter pylori* eradication [128,129]. Non-invasive techniques can also be used, in combination with a gradient-boosting decision tree, to build a predictive model identifying patients with GEA [130]. Other studies proposed an ML-based approach to identify patients who would require early readmission after surgical intervention of GEA [131].

2.2.2. Endoscopy-Based Approaches

Several studies trained CNNs to aid early detection in GEA, recently summarized in a meta-analysis that found superiority of applying DL for detection of Barrett's esophagus [132–137]. Currently, clinical trials are already investigating its sensitivity and specificity, if applied in a clinical setting, with several studies showing DL models to identify early GEA [138,139]. In detail, 3D endoscopy imaging techniques, in combination with DL,

may be applied to quantify the depth of Barrett's esophagus [140]. Of clinical relevance, another study found spectral endoscopy combined with DL more sensitive and specific to detect dysplastic vs. non-dysplastic Barrett than previous techniques [141]. Similarly, DL models have been used for the detection of intramucosal GEA using [142].

To identify individual patients and potential risk factors for recurrence of GEA after surgical intervention, Zhou et al., applied several regression/classification models and identified clinical variables that are associated with an increased risk [143]. A recent meta-analysis of several studies in Asian populations found that endoscopic imaging may also be analyzed to detect the presence of *Helicobacter pylori* infection [144].

2.2.3. Genomic-Based Approaches

Several studies used gene expression signatures, with array techniques or using NanoString, in addition to other techniques of RNA sequencing, to identify patients who would respond to chemotherapy in GEA [145]. In parallel, Chen et al., proposed seven immune-related genes to predict prognosis in GEA by applying a regression analysis [146]. Moreover, supportive vector machines have been applied to identify novel markers from circulating tumor cell-free DNA [147]. Recently, a multi-omics approach could identify responses to neoadjuvant therapy in GEA [148].

By following a complex combination of (similarity) clustering, Yuan et al., identified previously unrecognized non-coding long RNAs (lncRNAs) in gastric cancers [149]. In parallel, Li et al., compared several classification/regression models to identify novel lncRNAs in GEA [150]. Usually, a combination of both supervised and unsupervised techniques is used to identify subgroups of patients. For instance, to detect immunological subtypes of gastric cancer Chen et al., used a K-means clustering algorithm to detect subgroups based on RNA expression data and then trained a CNN to detect these subtypes using virtual-whole-slide images [120].

Genome-wide association studies identified novel susceptibility genes to gastric cancer using a random forest model [151]. By using several clustering algorithms of different sources of genomic data of 70 gastric cancer patients, Wang et al., proposed the detection of molecular subtypes in GEA [152]. After combing gene expression data and DNA methylation data for subsequent feature selection, Zhang et al., trained a model to detect novel biomarkers for discriminating between tumor and normal mucosa [153].

Owen et al., harvested mucosa tissue from different anatomical locations of the stomach to identify an overlap between Barrett mucosa and found an association to submucosal glands by single-cell RNA sequencing [154]. Here, and within other studies applying single-cell RNA sequencing, SC3 consensus clustering has been applied as an unsupervised learning method to allow the identification of certain genes that could distinguish common alterations in mucosa tissue [154,155].

2.2.4. Radiology-Based Approaches

Another example of ML applications in GEA is radiology, where different imaging modalities are used, most frequently CT imaging. For instance, CT imaging objects have been used to predict response to neoadjuvant therapy or to characterize tumor stromal components [156,157]. In a recent study, Lin et al., trained a CNN to detect lymph node metastasis by analyzing perioperative CT images of patients with gastric cancer. In addition, and relevant to potential therapeutic de-escalation therapy and patient surveillance, CT scans may also be used to monitor responses to (neoadjuvant) chemotherapy in GEA [158,159]. Other attempts involved training a model to aid the detection of GEA using CT scans [160].

Liu et al., followed an integrative approach of combining preoperative biomarkers including tissue biopsies, tumor markers, and CT image objects to predict lymph node metastasis in GEA by applying regression analysis and combined this to a multivariate model [161]. In parallel, similar image object information have been used to predict the risk

of peritoneal metastases using gradient boosting machines [162]. Others applied DL models to detect metastasis using CT image objects, in addition to adequate staging [163,164].

2.2.5. Digital Pathology and Virtual Microscopy-Based Approaches

Current examples that facilitate virtual whole slide images from regular H&E stains, that are generated within routine pathology workflow, include subtyping gastric cancer by convolutional neural networks [165,166]. In a recent study, Wang et al., trained a DL segmentation model to identify tumor regions within lymph nodes of gastric cancer patients and showed that this may serve as an interpretable independent prognostic factor in GEA [167]. In a study from our group, we developed a decision support system that combines morphological image operations to detect areas of relevance in large virtual whole-slide-image objects and proposes areas of *Helicobacter pylori* presence that can increase the sensitivity of identifying HP in gastric cancer biopsies, both of standard H&E staining and specialized Giemsa staining [168].

Park et al., trained a DL algorithm to identify gastric cancers in endoscopy biopsy specimens and showed that the system can increase time to diagnosis and may be potentially applied in countries with a lack of specialized pathologists [169]. Similarly, a recent multicentric study built a DL-based algorithm to aid in the diagnosis of gastric cancer and applied this using data from different scanners and different hospitals showing its generalization [170]. Sali et al., compared supervised and unsupervised DL-based models to identify dysplastic and non-dysplastic Barrett's Esophagus by analyzing virtual whole-slide images and found that unsupervised models achieved better results in comparison to supervised DL [171]. To reveal potential prognostic biomarkers of the TME in GEA, Meier et al., applied a DL model using tissue microarrays of a Japanese cohort [172]. Recent articles summarized potential requirements to more widely applying DL in gastrointestinal pathology [173,174], in addition to a systematic review highlighting applications of virtual whole-slide image analysis in GEA [175].

A retrospective multicentric study by Muti et al., built and validated a DL model predicting microsatellite and Epstein–Barr virus-associated GEA subtypes within a cohort of more than 2500 patients using scanned H&E whole-slide-images [176]. Although these advances appear to align with a recent success story of applying DL on histological images to predict microsatellite instability within colorectal cancer, published by the same group, it remains to be determined whether a molecular classification using DL would add benefit to the treatment of patients within prospective multicentric trials [177]. However, these proof-of-concept studies clearly indicate that molecular phenotyping using histological images may be of clinical interest. Future trials need to determine the exact value of these techniques as screening or eventually as additional parameters.

Broad and deep genetic sequencing efforts of tumor tissues and additional molecular analyses by "histological genotyping" could provide biomarkers of response but identify new targets for the treatment of a given patient. However, is it possible, for instance, that patients classified as microsatellite stable with help of DL applied on histological images show resistance to ICI despite the suggestion of response by genetic-based classification? Could we identify patients more likely to respond to ICI by building DL models end-to-end for treatment outcome as an alternative to more cumbersome molecular subtyping?

2.3. Current Status, Future Directions and Challanges of ML in GEA

2.3.1. Current Status of Machine Learning

Different ML models and techniques have been applied by several medical disciplines for object detection, segmentation, classification, and prognostic modeling using structured and unstructured data (Figures 2 and 3). Many of these applications appear to operate parallel to biomarkers that are already established. For instance, while the detection of molecular alterations using imaging objects may reduce costs and can potentially save time, there are already established techniques with approved drugs and sufficient sensitivity to allow identification of patients that will qualify for targeted therapies [92].

However, given the complexity of human cancers and their evolution during progression, where therapeutic pressure can select for drug-resistant clones and alter the TME, it would be of great relevance to use ML models to provide a more integrative view of the TME and its dynamic changes (Figure 1) [178–180].

2.3.2. Challenges and Future Directions

In addition to collecting tumor biopsies from cancer patients during treatment, an approach requiring invasive surgical procedures that may not always be feasible, radiological imaging data, and blood biomarkers may be used to gain more information on the TME of GEA from different time points during disease progression and therapy [181–183]. This would initially require finding surrogate markers or to generate models that find correlates of different TME phenotypes from these (preferable non-invasive) measures. For instance, circulating T-cell might be used to understand mechanisms of immune evasion, in addition to cytokine in blood circulation [184]. We and others have studied angiogenic biomarkers that may be useful biomarkers for tumor vascularization and vascular function, to facilitate the use of immunotherapy and anti-angiogenic therapies alone or in combination [56,185,186].

In the future, combined omics approaches that integrate most data resources will be gathered to retrieve biomarkers using ML models that will add a more holistic view on cancer progression, treatment resistance, and therefore optimal therapeutic decision making. Despite single diagnostic modalities that are revealing diagnostic or therapeutic proof of concept, this will unleash the full potential of well-annotated and well characterized datasets providing clinicians with the necessary information for decision making during the management of GEAs (Figure 4).
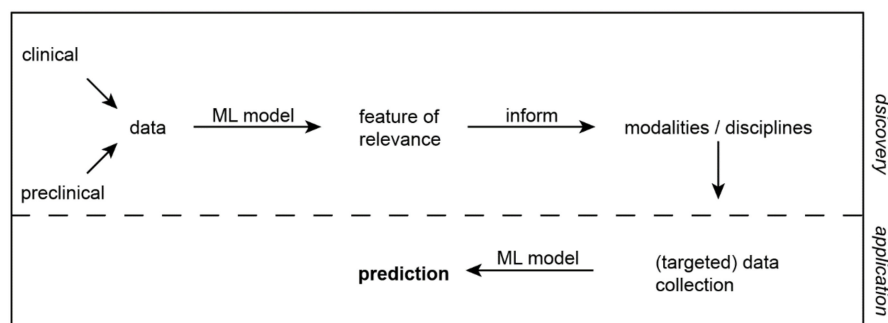
Figure 4. Future directions of machine learning in GEA. Within the discovery phase of machine learning, multi-omics approaches that are collecting data of various sources, including RNA and DNA sequencing, epigenetics, proteomics, imaging data, and metabolics will help to understand the TME, especially considering dynamic changes of cancer progression and treatment. These data will be integrated using machine learning and relevant information from all disciplines/modalities will be used to then inform the specialties what information (features) are necessary to apply specialized machine learning models that will predict individual disease traits, such as response to therapy and individual treatment strategies.

Here, modern technologies may be used to decipher aspects of the TME providing information that are not accessible by other technologies. In detail, in a first discovery phase (Figure 4), application of several omics technologies in parallel will be necessary to identify molecular and spatial characteristics of GEA that help to identify subtypes of the TME aiding dedicated therapeutic approaches. While ML models will be used during this phase to discover these characteristics, unsupervised algorithms that allow the detection of relevant biomarkers will be necessary to guide diagnostic modalities in the application phase.

Although ML, and in particular the application of DL for unstructured imaging data, could increase the sensitivity and specificity of cancer diagnosis, with detection of molecular subtypes, identification of subgroups, and stratification of patients, these advances need

to be validated and certified by regulatory bodies [187]. While many of the mentioned studies supplied proof of concept, a combination of biomarkers would require dedicated development for companion diagnostic applications.

At the same time, some of the mentioned models act as a "black box" where it appears to be difficult to decide what a given model detects, although reverse engineering, and study design, may allow for understanding the decisions of a network [188–190]. Interestingly, one may argue that increasing accuracy may be more important than understanding every aspect of a given model, especially given intra- and interobserver variability in medical decision making, in line with other examples of uncertainty, including mechanisms of drug action and disease mechanisms in medicine [191]. Other challenges that need to be overcome [192] include generating, validating, and applying models that can account for missing data points [193], highlighting the need for adequate preprocessing of data, in addition to normalization methods accounting for data variation [194].

To this end, medical disciplines need to be trained properly to understand the limitations of ML models. Likely, with the development of different DL architectures and ML techniques these applications need to undergo constant changes and adaption, requiring trained personal to address these challenges.

2.3.3. Summary

In summary, the particularities of the TME of GEA need to be defined in a dynamic fashion to aid the current applications of ML in this cancer entity. This will only be possible if data from different disciplines are combined, aiming to gather relevant information that may inform therapeutic decisions. Here, we summarized the current understanding in different medical specialties and discussed the challenges that need to be overcome to provide a more integrative view of the TME of GEA and facilitate clinical translation for the improvement of personalized therapies in this aggressive malignancy.

3. Conclusions

Clearly, the ML field is still in its infancy and is focused primarily on discovery and proof of concept studies, but there is promise that the translation phase of ML is within the immediate future.

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References

1. Egeblad, M.; Nakasone, E.S.; Werb, Z. Tumors as Organs: Complex Tissues that Interface with the Entire Organism. *Dev. Cell* **2010**, *18*, 884–901. [CrossRef]
2. Spranger, S.; Bao, R.; Gajewski, T.F. Melanoma-intrinsic β-catenin signalling prevents anti-tumour immunity. *Nature* **2015**, *523*, 231–235. [CrossRef] [PubMed]
3. Fearon, E.R.; Vogelstein, B. A genetic model for colorectal tumorigenesis. *Cell* **1990**, *61*, 759–767. [CrossRef]

4. Harlin, H.; Meng, Y.; Peterson, A.C.; Zha, Y.; Tretiakova, M.; Slingluff, C.; McKee, M.; Gajewski, T.F. Chemokine expression in melanoma metastases associated with CD8+ T-cell recruitment. *Cancer Res.* **2009**, *69*, 3077–3085. [CrossRef] [PubMed]

5. Briukhovetska, D.; Dörr, J.; Endres, S.; Libby, P.; Dinarello, C.A.; Kobold, S. Interleukins in cancer: From biology to therapy. *Nat. Rev. Cancer* **2021**, *21*, 481–499. [CrossRef] [PubMed]

6. Dranoff, G. Cytokines in cancer pathogenesis and cancer therapy. *Nat. Rev. Cancer* **2004**, *4*, 11–22. [CrossRef]

7. Tomlinson, I.P.; Novelli, M.R.; Bodmer, W.F. The mutation rate and cancer. *Proc. Natl. Acad. Sci. USA* **1996**, *93*, 14800–14803. [CrossRef] [PubMed]

8. Goel, S.; Duda, D.G.; Xu, L.; Munn, L.L.; Boucher, Y.; Fukumura, D.; Jain, R.K. Normalization of the Vasculature for Treatment of Cancer and Other Diseases. *Physiol. Rev.* **2011**, *91*, 1071–1121. [CrossRef]

9. Jain, R.K. Antiangiogenesis Strategies Revisited: From Starving Tumors to Alleviating Hypoxia. *Cancer Cell* **2014**, *26*, 605–622. [CrossRef]

10. Vitale, I.; Shema, E.; Loi, S.; Galluzzi, L. Intratumoral heterogeneity in cancer progression and response to immunotherapy. *Nat. Med.* **2021**, *27*, 212–224. [CrossRef]

11. McGranahan, N.; Swanton, C. Clonal Heterogeneity and Tumor Evolution: Past, Present, and the Future. *Cell* **2017**, *168*, 613–628. [CrossRef]

12. Kalluri, R.; Weinberg, R.A. The basics of epithelial-mesenchymal transition. *J. Clin. Investig.* **2009**, *119*, 1420–1428. [CrossRef] [PubMed]

13. Hanahan, D.; Weinberg, R.A. Hallmarks of Cancer: The Next Generation. *Cell* **2011**, *144*, 646–674. [CrossRef]

14. Lehmann, B.; Biburger, M.; Brückner, C.; Ipsen-Escobedo, A.; Gordan, S.; Lehmann, C.; Voehringer, D.; Winkler, T.; Schaft, N.; Dudziak, D.; et al. Tumor location determines tissue-specific recruitment of tumor-associated macrophages and antibody-dependent immunotherapy response. *Sci. Immunol.* **2017**, *2*, eaah6413. [CrossRef]

15. Sung, H.; Ferlay, J.; Siegel, R.L.; Laversanne, M.; Soerjomataram, I.; Jemal, A.; Bray, F. Global Cancer Statistics 2020: GLOBOCAN Estimates of Incidence and Mortality Worldwide for 36 Cancers in 185 Countries. *CA Cancer J. Clin.* **2021**, *71*, 209–249. [CrossRef]

16. Rustgi, A.K.; El-Serag, H.B. Esophageal Carcinoma. *N. Engl. J. Med.* **2014**, *371*, 2499–2509. [CrossRef] [PubMed]

17. Pennathur, A.; Gibson, M.K.; Jobe, B.A.; Luketich, J.D. Oesophageal carcinoma. *Lancet* **2013**, *381*, 400–412. [CrossRef]

18. Wild, C.P.; Hardie, L.J. Reflux, Barrett's oesophagus and adenocarcinoma: Burning questions. *Nat. Rev. Cancer* **2003**, *3*, 676–684. [CrossRef]

19. Ajani, J.A.; Lee, J.; Sano, T.; Janjigian, Y.Y.; Fan, D.; Song, S. Gastric adenocarcinoma. *Nat. Rev. Dis. Prim.* **2017**, *3*, 17036. [CrossRef]

20. Oliveira, C.; Seruca, R.; Carneiro, F. Hereditary gastric cancer. *Best Pract. Res. Clin. Gastroenterol.* **2009**, *23*, 147–157. [CrossRef]

21. Bass, A.J.; Thorsson, V.; Shmulevich, I.; Reynolds, S.M.; Miller, M.; Bernard, B.; Hinoue, T.; Laird, P.W.; Curtis, C.; Shen, H.; et al. Comprehensive molecular characterization of gastric adenocarcinoma. *Nature* **2014**, *513*, 202–209. [CrossRef]

22. Nagaraja, A.K.; Kikuchi, O.; Bass, A.J. Genomics and Targeted Therapies in Gastroesophageal Adenocarcinoma. *Cancer Discov.* **2019**, *9*, 1656–1672. [CrossRef] [PubMed]

23. Cancer Genome Atlas Research Network. Integrated genomic characterization of oesophageal carcinoma. *Nature* **2017**, *541*, 169. [CrossRef]

24. Greally, M.; Agarwal, R.; Ilson, D.H. Optimal management of gastroesophageal junction cancer. *Cancer* **2019**, *125*, 1990–2001. [CrossRef] [PubMed]

25. Dulak, A.M.; Stojanov, P.; Peng, S.; Lawrence, M.S.; Fox, C.; Stewart, C.; Bandla, S.; Imamura, Y.; Schumacher, S.E.; Shefler, E.; et al. Exome and whole-genome sequencing of esophageal adenocarcinoma identifies recurrent driver events and mutational complexity. *Nat. Genet.* **2013**, *45*, 478–486. [CrossRef] [PubMed]

26. Blank, C.; Gajewski, T.F.; Mackensen, A. Interaction of PD-L1 on tumor cells with PD-1 on tumor-specific T cells as a mechanism of immune evasion: Implications for tumor immunotherapy. *Cancer Immunol. Immunother.* **2005**, *54*, 307–314. [CrossRef] [PubMed]

27. Patel, S.P.; Kurzrock, R. PD-L1 Expression as a Predictive Biomarker in Cancer Immunotherapy. *Mol. Cancer Ther.* **2015**, *14*, 847–856. [CrossRef]

28. Taube, J.M.; Klein, A.; Brahmer, J.R.; Xu, H.; Pan, X.; Kim, J.H.; Chen, L.; Pardoll, D.M.; Topalian, S.L.; Anders, R.A. Association of PD-1, PD-1 Ligands, and Other Features of the Tumor Immune Microenvironment with Response to Anti–PD-1 Therapy. *Clin. Cancer Res.* **2014**, *20*, 5064–5074. [CrossRef] [PubMed]

29. Larkin, J.; Hodi, F.S.; Wolchok, J.D. Combined Nivolumab and Ipilimumab or Monotherapy in Untreated Melanoma. *N. Engl. J. Med.* **2015**, *373*, 1270–1271. [CrossRef] [PubMed]

30. Daud, A.I.; Wolchok, J.D.; Robert, C.; Hwu, W.-J.; Weber, J.S.; Ribas, A.; Hodi, F.S.; Joshua, A.M.; Kefford, R.; Hersey, P.; et al. Programmed Death-Ligand 1 Expression and Response to the Anti–Programmed Death 1 Antibody Pembrolizumab in Melanoma. *J. Clin. Oncol.* **2016**, *34*, 4102–4109. [CrossRef]

31. Ferrara, N. Vascular endothelial growth factor: Basic science and clinical progress. *Endocr. Rev.* **2004**, *25*, 581–611. [CrossRef]

32. Kalluri, R.; Zeisberg, M. Fibroblasts in cancer. *Nat. Rev. Cancer* **2006**, *6*, 392–401. [CrossRef]

33. Kobayashi, H.; Enomoto, A.; Woods, S.L.; Burt, A.D.; Takahashi, M.; Worthley, D.L. Cancer-associated fibroblasts in gastrointestinal cancer. *Nat. Rev. Gastroenterol. Hepatol.* **2019**, *16*, 282–295. [CrossRef]

34. Witz, I.P. The tumor microenvironment: The making of a paradigm. *Cancer Microenviron.* **2009**, *2* (Suppl. S1), 9–17. [CrossRef]

35. Whiteside, T.L. The tumor microenvironment and its role in promoting tumor growth. *Oncogene* **2008**, *27*, 5904–5912. [CrossRef] [PubMed]

36. Noy, R.; Pollard, J.W. Tumor-associated macrophages: From mechanisms to therapy. *Immunity* **2014**, *41*, 49–61. [CrossRef] [PubMed]
37. Engblom, C.; Pfirschke, C.; Pittet, M.J. The role of myeloid cells in cancer therapies. *Nat. Rev. Cancer* **2016**, *16*, 447–462. [CrossRef]
38. Catenacci, D.V.T.; Tebbutt, N.C.; Davidenko, I.; Murad, A.M.; Al-Batran, S.E.; Ilson, D.H.; Tjulandin, S.; Gotovkin, E.; Karaszewska, B.; Bondarenko, I.; et al. Rilotumumab plus epirubicin, cisplatin, and capecitabine as first-line therapy in advanced MET-positive gastric or gastro-oesophageal junction cancer (RILOMET-1): A randomised, double-blind, placebo-controlled, phase 3 trial. *Lancet Oncol.* **2017**, *18*, 1467–1482. [CrossRef]
39. Wilke, H.; Muro, K.; Van Cutsem, E.; Oh, S.C.; Bodoky, G.; Shimada, Y.; Hironaka, S.; Sugimoto, N.; Lipatov, O.; Kim, T.Y.; et al. Ramucirumab plus paclitaxel versus placebo plus paclitaxel in patients with previously treated advanced gastric or gastro-oesophageal junction adenocarcinoma (RAINBOW): A double-blind, randomised phase 3 trial. *Lancet Oncol.* **2014**, *15*, 1224–1235. [CrossRef]
40. Fuchs, C.S.; Tomasek, J.; Yong, C.J.; Dumitru, F.; Passalacqua, R.; Goswami, C.; Safran, H.; dos Santos, L.V.; Aprile, G.; Ferry, D.R.; et al. Ramucirumab monotherapy for previously treated advanced gastric or gastro-oesophageal junction adenocarcinoma (REGARD): An international, randomised, multicentre, placebo-controlled, phase 3 trial. *Lancet* **2014**, *383*, 31–39. [CrossRef]
41. Chan, D.L.; Sjoquist, K.M.; Goldstein, D.; Price, T.J.; Martin, A.J.; Bang, Y.J.; Kang, Y.K.; Pavlakis, N. The effect of anti-angiogenic agents on overall survival in metastatic oesophago-gastric cancer: A systematic review and meta-analysis. *PLoS ONE* **2017**, *12*, e0172307. [CrossRef]
42. Li, J.; Qin, S.; Xu, J.; Xiong, J.; Wu, C.; Bai, Y.; Liu, W.; Tong, J.; Liu, Y.; Xu, R.; et al. Randomized, Double-Blind, Placebo-Controlled Phase III Trial of Apatinib in Patients with Chemotherapy-Refractory Advanced or Metastatic Adenocarcinoma of the Stomach or Gastroesophageal Junction. *J. Clin. Oncol.* **2016**, *34*, 1448–1454. [CrossRef]
43. Keir, M.E.; Butte, M.J.; Freeman, G.J.; Sharpe, A.H. PD-1 and its ligands in tolerance and immunity. *Annu. Rev. Immunol.* **2008**, *26*, 677–704. [CrossRef] [PubMed]
44. Janjigian, Y.Y.; Bendell, J.; Calvo, E.; Kim, J.W.; Ascierto, P.A.; Sharma, P.; Ott, P.A.; Peltola, K.; Jaeger, D.; Evans, J.; et al. CheckMate-032 Study: Efficacy and Safety of Nivolumab and Nivolumab Plus Ipilimumab in Patients with Metastatic Esophagogastric Cancer. *J. Clin. Oncol.* **2018**, *36*, 2836–2844. [CrossRef] [PubMed]
45. Janjigian, Y.Y.; Bendell, J.C.; Calvo, E.; Kim, J.W.; Ascierto, P.A.; Sharma, P.; Ott, P.A.; Bono, P.; Jaeger, D.; Evans, T.R.J.; et al. CheckMate-032: Phase I/II, open-label study of safety and activity of nivolumab (nivo) alone or with ipilimumab (ipi) in advanced and metastatic (A/M) gastric cancer (GC). *J. Clin. Oncol.* **2016**, *34*, 4010. [CrossRef]
46. Shah, M.A.; Kojima, T.; Hochhauser, D.; Enzinger, P.; Raimbourg, J.; Hollebecque, A.; Lordick, F.; Kim, S.-B.; Tajika, M.; Kim, H.T.; et al. Efficacy and Safety of Pembrolizumab for Heavily Pretreated Patients with Advanced, Metastatic Adenocarcinoma or Squamous Cell Carcinoma of the Esophagus: The Phase 2 KEYNOTE-180 Study. *JAMA Oncol.* **2019**, *5*, 546–550. [CrossRef] [PubMed]
47. Kang, Y.-K.; Boku, N.; Satoh, T.; Ryu, M.-H.; Chao, Y.; Kato, K.; Chung, H.C.; Chen, J.-S.; Muro, K.; Kang, W.K.; et al. Nivolumab in patients with advanced gastric or gastro-oesophageal junction cancer refractory to, or intolerant of, at least two previous chemotherapy regimens (ONO-4538-12, ATTRACTION-2): A randomised, double-blind, placebo-controlled, phase 3 trial. *Lancet* **2017**, *390*, 2461–2471. [CrossRef]
48. Fuchs, C.S.; Doi, T.; Jang, R.W.; Muro, K.; Satoh, T.; Machado, M.; Sun, W.; Jalal, S.I.; Shah, M.A.; Metges, J.P.; et al. Safety and Efficacy of Pembrolizumab Monotherapy in Patients with Previously Treated Advanced Gastric and Gastroesophageal Junction Cancer: Phase 2 Clinical KEYNOTE-059 Trial. *JAMA Oncol.* **2018**, *4*, e180013. [CrossRef]
49. Bang, Y.J.; Kang, Y.K.; Catenacci, D.V.; Muro, K.; Fuchs, C.S.; Geva, R.; Hara, H.; Golan, T.; Garrido, M.; Jalal, S.I.; et al. Pembrolizumab alone or in combination with chemotherapy as first-line therapy for patients with advanced gastric or gastro-sophageal junction adenocarcinoma: Results from the phase II nonrandomized KEYNOTE-059 study. *Gastric Cancer* **2019**, *22*, 828–837. [CrossRef] [PubMed]
50. Twomey, J.D.; Zhang, B. Cancer Immunotherapy Update: FDA-Approved Checkpoint Inhibitors and Companion Diagnostics. *AAPS J.* **2021**, *23*, 39. [CrossRef]
51. Saeed, A.; Park, R.; Sun, W. The integration of immune checkpoint inhibitors with VEGF targeted agents in advanced gastric and gastroesophageal adenocarcinoma: A review on the rationale and results of early phase trials. *J. Hematol. Oncol.* **2021**, *14*, 13. [CrossRef] [PubMed]
52. Shigeta, K.; Matsui, A.; Kikuchi, H.; Klein, S.; Mamessier, E.; Chen, I.X.; Aoki, S.; Kitahara, S.; Inoue, K.; Shigeta, A.; et al. Regorafenib combined with PD1 blockade increases CD8 T-cell infiltration by inducing CXCL10 expression in hepatocellular carcinoma. *J. Immunother. Cancer* **2020**, *8*, e001435. [CrossRef] [PubMed]
53. Shigeta, K.; Datta, M.; Hato, T.; Kitahara, S.; Chen, I.X.; Matsui, A.; Kikuchi, H.; Mamessier, E.; Aoki, S.; Ramjiawan, R.R.; et al. Dual Programmed Death Receptor-1 and Vascular Endothelial Growth Factor Receptor-2 Blockade Promotes Vascular Normalization and Enhances Antitumor Immune Responses in Hepatocellular Carcinoma. *Hepatology* **2020**, *71*, 1247–1261. [CrossRef] [PubMed]
54. Ramjiawan, R.R.; Griffioen, A.W.; Duda, D.G. Anti-angiogenesis for cancer revisited: Is there a role for combinations with immunotherapy? *Angiogenesis* **2017**, *20*, 185–204. [CrossRef]

55. Meder, L.; Schuldt, P.; Thelen, M.; Schmitt, A.; Dietlein, F.; Klein, S.; Borchmann, S.; Wennhold, K.; Vlasic, I.; Oberbeck, S.; et al. Combined VEGF and PD-L1 Blockade Displays Synergistic Treatment Effects in an Autochthonous Mouse Model of Small Cell Lung Cancer. *Cancer Res.* **2018**, *78*, 4270–4281. [CrossRef]

56. Fukumura, D.; Kloepper, J.; Amoozgar, Z.; Duda, D.G.; Jain, R.K. Enhancing cancer immunotherapy using antiangiogenics: Opportunities and challenges. *Nat. Rev. Clin. Oncol.* **2018**, *15*, 325–340. [CrossRef]

57. Fukuoka, S.; Hara, H.; Takahashi, N.; Kojima, T.; Kawazoe, A.; Asayama, M.; Yoshii, T.; Kotani, D.; Tamura, H.; Mikamoto, Y.; et al. Regorafenib Plus Nivolumab in Patients with Advanced Gastric or Colorectal Cancer: An Open-Label, Dose-Escalation, and Dose-Expansion Phase Ib Trial (REGONIVO, EPOC1603). *J. Clin. Oncol.* **2020**, *38*, 2053–2061. [CrossRef]

58. Kauer, W.K.; Peters, J.H.; DeMeester, T.R.; Ireland, A.P.; Bremner, C.G.; Hagen, J.A. Mixed reflux of gastric and duodenal juices is more harmful to the esophagus than gastric juice alone. The need for surgical therapy re-emphasized. *Ann. Surg.* **1995**, *222*, 525–531. [CrossRef]

59. O'Riordan, J.M.; Abdel-latif, M.M.; Ravi, N.; McNamara, D.; Byrne, P.J.; McDonald, G.S.; Keeling, P.W.; Kelleher, D.; Reynolds, J.V. Proinflammatory cytokine and nuclear factor kappa-B expression along the inflammation-metaplasia-dysplasia-adenocarcinoma sequence in the esophagus. *Am. J. Gastroenterol.* **2005**, *100*, 1257–1264. [CrossRef]

60. Avidan, B.; Sonnenberg, A.; Schnell, T.G.; Chejfec, G.; Metz, A.; Sontag, S.J. Hiatal hernia size, Barrett's length, and severity of acid reflux are all risk factors for esophageal adenocarcinoma. *Am. J. Gastroenterol.* **2002**, *97*, 1930–1936. [CrossRef]

61. Fox, J.G.; Wang, T.C. Inflammation, atrophy, and gastric cancer. *J. Clin. Investig.* **2007**, *117*, 60–69. [CrossRef]

62. Coussens, L.M.; Werb, Z. Inflammation and cancer. *Nature* **2002**, *420*, 860–867. [CrossRef]

63. Eder, K.; Baffy, N.; Falus, A.; Fulop, A.K. The major inflammatory mediator interleukin-6 and obesity. *Inflamm. Res.* **2009**, *58*, 727–736. [CrossRef] [PubMed]

64. Park, H.S.; Park, J.Y.; Yu, R. Relationship of obesity and visceral adiposity with serum concentrations of CRP, TNF-alpha and IL-6. *Diabetes Res. Clin. Pract.* **2005**, *69*, 29–35. [CrossRef]

65. Incio, J.; Liu, H.; Suboj, P.; Chin, S.M.; Chen, I.X.; Pinter, M.; Ng, M.R.; Nia, H.T.; Grahovac, J.; Kao, S.; et al. Obesity-Induced Inflammation and Desmoplasia Promote Pancreatic Cancer Progression and Resistance to Chemotherapy. *Cancer Discov.* **2016**, *6*, 852–869. [CrossRef] [PubMed]

66. Ostrand-Rosenberg, S.; Sinha, P. Myeloid-derived suppressor cells: Linking inflammation and cancer. *J. Immunol.* **2009**, *182*, 4499–4506. [CrossRef]

67. Landskron, G.; De la Fuente, M.; Thuwajit, P.; Thuwajit, C.; Hermoso, M.A. Chronic Inflammation and Cytokines in the Tumor Microenvironment. *J. Immunol. Res.* **2014**, *2014*, 149185. [CrossRef]

68. Nagaraj, S.; Schrum, A.G.; Cho, H.I.; Celis, E.; Gabrilovich, D.I. Mechanism of T cell tolerance induced by myeloid-derived suppressor cells. *J. Immunol.* **2010**, *184*, 3106–3116. [CrossRef]

69. Nielsen, S.R.; Schmid, M.C. Macrophages as Key Drivers of Cancer Progression and Metastasis. *Mediat. Inflamm.* **2017**, *2017*, 9624760. [CrossRef]

70. Poh, A.R.; Ernst, M. Targeting Macrophages in Cancer: From Bench to Bedside. *Front. Oncol.* **2018**, *8*, 49. [CrossRef] [PubMed]

71. Tang, P.M.; Nikolic-Paterson, D.J.; Lan, H.Y. Macrophages: Versatile players in renal inflammation and fibrosis. *Nat. Rev. Nephrol.* **2019**, *15*, 144–158. [CrossRef] [PubMed]

72. Wang, J.; Li, D.; Cang, H.; Guo, B. Crosstalk between cancer and immune cells: Role of tumor-associated macrophages in the tumor microenvironment. *Cancer Med.* **2019**, *8*, 4709–4721. [CrossRef]

73. De Vries, N.L.; Mahfouz, A.; Koning, F.; de Miranda, N. Unraveling the Complexity of the Cancer Microenvironment with Multidimensional Genomic and Cytometric Technologies. *Front. Oncol.* **2020**, *10*, 1254. [CrossRef] [PubMed]

74. Xue, V.W.; Chung, J.Y.; Córdoba, C.A.G.; Cheung, A.H.; Kang, W.; Lam, E.W.; Leung, K.T.; To, K.F.; Lan, H.Y.; Tang, P.M. Transforming Growth Factor-β: A Multifunctional Regulator of Cancer Immunity. *Cancers* **2020**, *12*, 3099. [CrossRef]

75. Chung, J.Y.; Chan, M.K.; Li, J.S.; Chan, A.S.; Tang, P.C.; Leung, K.T.; To, K.F.; Lan, H.Y.; Tang, P.M. TGF-β Signaling: From Tissue Fibrosis to Tumor Microenvironment. *Int. J. Mol. Sci.* **2021**, *22*, 7575. [CrossRef] [PubMed]

76. Liu, B.; Zhou, M.; Li, X.; Zhang, X.; Wang, Q.; Liu, L.; Yang, M.; Yang, D.; Guo, Y.; Zhang, Q.; et al. Interrogation of gender disparity uncovers androgen receptor as the transcriptional activator for oncogenic miR-125b in gastric cancer. *Cell Death Dis.* **2021**, *12*, 441. [CrossRef] [PubMed]

77. Quaas, A.; Pamuk, A.; Klein, S.; Quantius, J.; Rehkaemper, J.; Barutcu, A.G.; Rueschoff, J.; Zander, T.; Gebauer, F.; Hillmer, A.; et al. Sex-specific prognostic effect of CD66b-positive tumor-infiltrating neutrophils (TANs) in gastric and esophageal adenocarcinoma. *Gastric Cancer* **2021**. [CrossRef]

78. Clausen, F.; Behrens, H.-M.; Krüger, S.; Röcken, C. Sexual dimorphism in gastric cancer: Tumor-associated neutrophils predict patient outcome only for women. *J. Cancer Res. Clin. Oncol.* **2020**, *146*, 53–66. [CrossRef]

79. Li, C.H.; Haider, S.; Shiah, Y.J.; Thai, K.; Boutros, P.C. Sex Differences in Cancer Driver Genes and Biomarkers. *Cancer Res.* **2018**, *78*, 5527–5537. [CrossRef]

80. Mathieu, L.N.; Kanarek, N.F.; Tsai, H.-L.; Rudin, C.M.; Brock, M.V. Age and sex differences in the incidence of esophageal adenocarcinoma: Results from the Surveillance, Epidemiology, and End Results (Seer) Registry (1973–2008). *Dis. Esophagus* **2014**, *27*, 757–763. [CrossRef] [PubMed]

81. Kim, H.-I.; Lim, H.; Moon, A. Sex Differences in Cancer: Epidemiology, Genetics and Therapy. *Biomol. Ther.* **2018**, *26*, 335–342. [CrossRef]

82. Setia, N.; Agoston, A.T.; Han, H.S.; Mullen, J.T.; Duda, D.G.; Clark, J.W.; Deshpande, V.; Mino-Kenudson, M.; Srivastava, A.; Lennerz, J.K.; et al. A protein and mRNA expression-based classification of gastric cancer. *Mod. Pathol.* **2016**, *29*, 772–784. [CrossRef]

83. Lei, Z.; Tan, I.B.; Das, K.; Deng, N.; Zouridis, H.; Pattison, S.; Chua, C.; Feng, Z.; Guan, Y.K.; Ooi, C.H.; et al. Identification of molecular subtypes of gastric cancer with different responses to PI3-kinase inhibitors and 5-fluorouracil. *Gastroenterology* **2013**, *145*, 554–565. [CrossRef]

84. Cristescu, R.; Lee, J.; Nebozhyn, M.; Kim, K.M.; Ting, J.C.; Wong, S.S.; Liu, J.; Yue, Y.G.; Wang, J.; Yu, K.; et al. Molecular analysis of gastric cancer identifies subtypes associated with distinct clinical outcomes. *Nat. Med.* **2015**, *21*, 449–456. [CrossRef]

85. Rodriquenz, M.G.; Roviello, G.; D'Angelo, A.; Lavacchi, D.; Roviello, F.; Polom, K. MSI and EBV Positive Gastric Cancer's Subgroups and Their Link with Novel Immunotherapy. *J. Clin. Med.* **2020**, *9*, 1427. [CrossRef]

86. Chao, J.; Fuchs, C.S.; Shitara, K.; Tabernero, J.; Muro, K.; Van Cutsem, E.; Bang, Y.-J.; De Vita, F.; Landers, G.; Yen, C.-J.; et al. Assessment of Pembrolizumab Therapy for the Treatment of Microsatellite Instability–High Gastric or Gastroesophageal Junction Cancer Among Patients in the KEYNOTE-059, KEYNOTE-061, and KEYNOTE-062 Clinical Trials. *JAMA Oncol.* **2021**, *7*, 895–902. [CrossRef] [PubMed]

87. Huang, Z.-H.; Ma, X.-W.; Zhang, J.; Li, X.; Lai, N.-L.; Zhang, S.-X. Cetuximab for esophageal cancer: An updated meta-analysis of randomized controlled trials. *BMC Cancer* **2018**, *18*, 1170. [CrossRef] [PubMed]

88. Petty, R.D.; Dahle-Smith, A.; Stevenson, D.A.J.; Osborne, A.; Massie, D.; Clark, C.; Murray, G.I.; Dutton, S.J.; Roberts, C.; Chong, I.Y.; et al. Gefitinib and EGFR Gene Copy Number Aberrations in Esophageal Cancer. *J. Clin. Oncol.* **2017**, *35*, 2279–2287. [CrossRef]

89. Xu, Y.; Xie, Z.; Shi, Y.; Zhang, M.; Pan, J.; Li, Y.; Lu, H. Gefitinib single drug in treatment of advanced esophageal cancer. *J. Cancer Res.* **2016**, *12*, C295–C297. [CrossRef]

90. Al-Kasspooles, M.; Moore, J.H.; Orringer, M.B.; Beer, D.G. Amplification and over-expression of the EGFR and erbB-2 genes in human esophageal adenocarcinomas. *Int. J. Cancer* **1993**, *54*, 213–219. [CrossRef]

91. Doi, T.; Shitara, K.; Naito, Y.; Shimomura, A.; Fujiwara, Y.; Yonemori, K.; Shimizu, C.; Shimoi, T.; Kuboki, Y.; Matsubara, N.; et al. Safety, pharmacokinetics, and antitumour activity of trastuzumab deruxtecan (DS-8201), a HER2-targeting antibody-drug conjugate, in patients with advanced breast and gastric or gastro-oesophageal tumours: A phase 1 dose-escalation study. *Lancet Oncol.* **2017**, *18*, 1512–1522. [CrossRef]

92. Van Cutsem, E.; Bang, Y.J.; Feng-Yi, F.; Xu, J.M.; Lee, K.W.; Jiao, S.C.; Chong, J.L.; López-Sanchez, R.I.; Price, T.; Gladkov, O.; et al. HER2 screening data from ToGA: Targeting HER2 in gastric and gastroesophageal junction cancer. *Gastric Cancer* **2015**, *18*, 476–484. [CrossRef]

93. Hecht, J.R.; Bang, Y.J.; Qin, S.K.; Chung, H.C.; Xu, J.M.; Park, J.O.; Jeziorski, K.; Shparyk, Y.; Hoff, P.M.; Sobrero, A.; et al. Lapatinib in Combination with Capecitabine Plus Oxaliplatin in Human Epidermal Growth Factor Receptor 2-Positive Advanced or Metastatic Gastric, Esophageal, or Gastroesophageal Adenocarcinoma: TRIO-013/LOGiC–A Randomized Phase III Trial. *J. Clin. Oncol.* **2016**, *34*, 443–451. [CrossRef]

94. Maher, S.G.; Gillham, C.M.; Duggan, S.P.; Smyth, P.C.; Miller, N.; Muldoon, C.; O'Byrne, K.J.; Sheils, O.M.; Hollywood, D.; Reynolds, J.V. Gene expression analysis of diagnostic biopsies predicts pathological response to neoadjuvant chemoradiotherapy of esophageal cancer. *Ann. Surg.* **2009**, *250*, 729–737. [CrossRef] [PubMed]

95. Luthra, R.; Wu, T.T.; Luthra, M.G.; Izzo, J.; Lopez-Alvarez, E.; Zhang, L.; Bailey, J.; Lee, J.H.; Bresalier, R.; Rashid, A.; et al. Gene expression profiling of localized esophageal carcinomas: Association with pathologic response to preoperative chemoradiation. *J. Clin. Oncol.* **2006**, *24*, 259–267. [CrossRef] [PubMed]

96. Schauer, M.; Janssen, K.P.; Rimkus, C.; Raggi, M.; Feith, M.; Friess, H.; Theisen, J. Microarray-based response prediction in esophageal adenocarcinoma. *Clin. Cancer Res.* **2010**, *16*, 330–337. [CrossRef]

97. Motoori, M.; Takemasa, I.; Yamasaki, M.; Komori, T.; Takeno, A.; Miyata, H.; Takiguchi, S.; Fujiwara, Y.; Yasuda, T.; Yano, M.; et al. Prediction of the response to chemotherapy in advanced esophageal cancer by gene expression profiling of biopsy samples. *Int. J. Oncol.* **2010**, *37*, 1113–1120. [CrossRef]

98. Duong, C.; Greenawalt, D.M.; Kowalczyk, A.; Ciavarella, M.L.; Raskutti, G.; Murray, W.K.; Phillips, W.A.; Thomas, R.J. Pretreatment gene expression profiles can be used to predict response to neoadjuvant chemoradiotherapy in esophageal cancer. *Ann. Surg. Oncol.* **2007**, *14*, 3602–3609. [CrossRef]

99. Oya, Y.; Hayakawa, Y.; Koike, K. Tumor microenvironment in gastric cancers. *Cancer Sci.* **2020**, *111*, 2696–2707. [CrossRef]

100. Lin, E.W.; Karakasheva, T.A.; Hicks, P.D.; Bass, A.J.; Rustgi, A.K. The tumor microenvironment in esophageal cancer. *Oncogene* **2016**, *35*, 5337–5349. [CrossRef] [PubMed]

101. Wang, F.; Wei, X.L.; Wang, F.H.; Xu, N.; Shen, L.; Dai, G.H.; Yuan, X.L.; Chen, Y.; Yang, S.J.; Shi, J.H.; et al. Safety, efficacy and tumor mutational burden as a biomarker of overall survival benefit in chemo-refractory gastric cancer treated with toripalimab, a PD-1 antibody in phase Ib/II clinical trial NCT02915432. *Ann. Oncol.* **2019**, *30*, 1479–1486. [CrossRef]

102. Yarchoan, M.; Hopkins, A.; Jaffee, E.M. Tumor Mutational Burden and Response Rate to PD-1 Inhibition. *N. Engl. J. Med.* **2017**, *377*, 2500–2501. [CrossRef]

103. Noh, M.-G.; Yoon, Y.; Kim, G.; Kim, H.; Lee, E.; Kim, Y.; Park, C.; Lee, K.-H.; Park, H. Practical prediction model of the clinical response to programmed death-ligand 1 inhibitors in advanced gastric cancer. *Exp. Mol. Med.* **2021**, *53*, 223–234. [CrossRef] [PubMed]

104. Lu, Z.; Chen, H.; Jiao, X.; Zhou, W.; Han, W.; Li, S.; Liu, C.; Gong, J.; Li, J.; Zhang, X.; et al. Prediction of immune checkpoint inhibition with immune oncology-related gene expression in gastrointestinal cancer using a machine learning classifier. *J. Immunother. Cancer* **2020**, *8*, e000631. [CrossRef]

105. Clemente, C.G.; Mihm, M.C., Jr.; Bufalino, R.; Zurrida, S.; Collini, P.; Cascinelli, N. Prognostic value of tumor infiltrating lymphocytes in the vertical growth phase of primary cutaneous melanoma. *Cancer* **1996**, *77*, 1303–1310. [CrossRef]

106. Klein, S.; Mauch, C.; Brinker, K.; Noh, K.-W.; Knez, S.; Büttner, R.; Quaas, A.; Helbig, D. Tumor infiltrating lymphocyte clusters are associated with response to immune checkpoint inhibition in BRAF V600E/K mutated malignant melanomas. *Sci. Rep.* **2021**, *11*, 1834. [CrossRef]

107. Zeng, D.; Wu, J.; Luo, H.; Li, Y.; Xiao, J.; Peng, J.; Ye, Z.; Zhou, R.; Yu, Y.; Wang, G.; et al. Tumor microenvironment evaluation promotes precise checkpoint immunotherapy of advanced gastric cancer. *J. Immunother. Cancer* **2021**, *9*, e002467. [CrossRef] [PubMed]

108. McGranahan, N.; Furness, A.J.S.; Rosenthal, R.; Ramskov, S.; Lyngaa, R.; Saini, S.K.; Jamal-Hanjani, M.; Wilson, G.A.; Birkbak, N.J.; Hiley, C.T.; et al. Clonal neoantigens elicit T cell immunoreactivity and sensitivity to immune checkpoint blockade. *Science* **2016**, *351*, 1463–1469. [CrossRef]

109. Kim, S.; Kim, H.S.; Kim, E.; Lee, M.G.; Shin, E.C.; Paik, S.; Kim, S. Neopepsee: Accurate genome-level prediction of neoantigens by harnessing sequence and amino acid immunogenicity information. *Ann. Oncol.* **2018**, *29*, 1030–1036. [CrossRef]

110. Maleki Vareki, S. High and low mutational burden tumors versus immunologically hot and cold tumors and response to immune checkpoint inhibitors. *J. Immunother. Cancer* **2018**, *6*, 157. [CrossRef]

111. Wang, P.; Chen, Y.; Wang, C. Beyond Tumor Mutation Burden: Tumor Neoantigen Burden as a Biomarker for Immunotherapy and Other Types of Therapy. *Front. Oncol.* **2021**, *11*, 672677. [CrossRef]

112. LeCun, Y.; Bengio, Y.; Hinton, G. Deep learning. *Nature* **2015**, *521*, 436–444. [CrossRef] [PubMed]

113. Tang, P.M.; Zhou, S.; Li, C.J.; Liao, J.; Xiao, J.; Wang, Q.M.; Lian, G.Y.; Li, J.; Huang, X.R.; To, K.F.; et al. The proto-oncogene tyrosine protein kinase Src is essential for macrophage-myofibroblast transition during renal scarring. *Kidney Int.* **2018**, *93*, 173–187. [CrossRef] [PubMed]

114. Tang, P.M.; Zhang, Y.Y.; Xiao, J.; Tang, P.C.; Chung, J.Y.; Li, J.; Xue, V.W.; Huang, X.R.; Chong, C.C.; Ng, C.F.; et al. Neural transcription factor Pou4f1 promotes renal fibrosis via macrophage-myofibroblast transition. *Proc. Natl. Acad. Sci. USA* **2020**, *117*, 20741–20752. [CrossRef]

115. Bushati, N.; Smith, J.; Briscoe, J.; Watkins, C. An intuitive graphical visualization technique for the interrogation of transcriptome data. *Nucleic Acids Res.* **2011**, *39*, 7380–7389. [CrossRef] [PubMed]

116. Li, W.; Cerise, J.E.; Yang, Y.; Han, H. Application of t-SNE to human genetic data. *J. Bioinform. Comput. Biol.* **2017**, *15*, 1750017. [CrossRef]

117. Kobak, D.; Berens, P. The art of using t-SNE for single-cell transcriptomics. *Nat. Commun.* **2019**, *10*, 5416. [CrossRef]

118. Islam, M.; Chen, B.; Spraggins, J.M.; Kelly, R.T.; Lau, K.S. Use of Single-Cell -Omic Technologies to Study the Gastrointestinal Tract and Diseases, From Single Cell Identities to Patient Features. *Gastroenterology* **2020**, *159*, 453–466.e451. [CrossRef]

119. Yoon, H.G.; Oh, D.; Noh, J.M.; Cho, W.K.; Sun, J.M.; Kim, H.K.; Zo, J.I.; Shim, Y.M.; Kim, K. Machine learning model for predicting excessive muscle loss during neoadjuvant chemoradiotherapy in oesophageal cancer. *J. Cachexia Sarcopenia Muscle* **2021**. [CrossRef]

120. Zhao, Z.; Cheng, X.; Sun, X.; Ma, S.; Feng, H.; Zhao, L. Prediction Model of Anastomotic Leakage Among Esophageal Cancer Patients After Receiving an Esophagectomy: Machine Learning Approach. *JMIR Med. Inf.* **2021**, *9*, e27110. [CrossRef]

121. Barragán-Montero, A.M.; Thomas, M.; Defraene, G.; Michiels, S.; Haustermans, K.; Lee, J.A.; Sterpin, E. Deep learning dose prediction for IMRT of esophageal cancer: The effect of data quality and quantity on model performance. *Phys. Med.* **2021**, *83*, 52–63. [CrossRef]

122. Xu, L.; Hu, J.; Song, Y.; Bai, S.; Yi, Z. Clinical target volume segmentation for stomach cancer by stochastic width deep neural network. *Med. Phys.* **2021**, *48*, 1720–1730. [CrossRef]

123. Jiao, S.X.; Wang, M.L.; Chen, L.X.; Liu, X.W. Evaluation of dose-volume histogram prediction for organ-at risk and planning target volume based on machine learning. *Sci. Rep.* **2021**, *11*, 3117. [CrossRef]

124. Zhu, J.; Chen, X.; Yang, B.; Bi, N.; Zhang, T.; Men, K.; Dai, J. Evaluation of Automatic Segmentation Model with Dosimetric Metrics for Radiotherapy of Esophageal Cancer. *Front. Oncol.* **2020**, *10*, 564737. [CrossRef] [PubMed]

125. Jiang, D.; Yan, H.; Chang, N.; Li, T.; Mao, R.; Du, C.; Guo, B.; Liu, J. Convolutional neural network-based dosimetry evaluation of esophageal radiation treatment planning. *Med. Phys.* **2020**, *47*, 4735–4742. [CrossRef] [PubMed]

126. Rahman, S.A.; Walker, R.C.; Maynard, N.; Trudgill, N.; Crosby, T.; Cromwell, D.A.; Underwood, T.J. The AUGIS Survival Predictor: Prediction of Long-term and Conditional Survival after Esophagectomy Using Random Survival Forests. *Ann. Surg.* **2021**. [CrossRef] [PubMed]

127. Aslam, M.A.; Xue, C.; Chen, Y.; Zhang, A.; Liu, M.; Wang, K.; Cui, D. Breath analysis based early gastric cancer classification from deep stacked sparse autoencoder neural network. *Sci. Rep.* **2021**, *11*, 4014. [CrossRef]

128. Leung, W.K.; Cheung, K.S.; Li, B.; Law, S.Y.K.; Lui, T.K.L. Applications of machine learning models in the prediction of gastric cancer risk in patients after Helicobacter pylori eradication. *Aliment. Pharm.* **2021**, *53*, 864–872. [CrossRef]

129. Chen, T.; Guestrin, C. XGBoost: A Scalable Tree Boosting System. In Proceedings of the 22nd ACM SIGKDD International Conference on Knowledge Discovery and Data Mining (KDD'16), San Francisco, CA, USA, 13–17 August 2016; pp. 785–794. [CrossRef]

130. Zhu, S.L.; Dong, J.; Zhang, C.; Huang, Y.B.; Pan, W. Application of machine learning in the diagnosis of gastric cancer based on noninvasive characteristics. *PLoS ONE* **2020**, *15*, e0244869. [CrossRef]

131. Bolourani, S.; Tayebi, M.A.; Diao, L.; Wang, P.; Patel, V.; Manetta, F.; Lee, P.C. Using machine learning to predict early readmission following esophagectomy. *J. Thorac. Cardiovasc. Surg.* **2021**, *161*, 1926–1939.e1928. [CrossRef]

132. Bhatti, K.M.; Khanzada, Z.S.; Kuzman, M.; Ali, S.M.; Iftikhar, S.Y.; Small, P. Diagnostic Performance of Artificial Intelligence-Based Models for the Detection of Early Esophageal Cancers in Barret's Esophagus: A Meta-Analysis of Patient-Based Studies. *Cureus* **2021**, *13*, e15447. [CrossRef]

133. Visaggi, P.; Barberio, B.; Ghisa, M.; Ribolsi, M.; Savarino, V.; Fassan, M.; Valmasoni, M.; Marchi, S.; de Bortoli, N.; Savarino, E. Modern Diagnosis of Early Esophageal Cancer: From Blood Biomarkers to Advanced Endoscopy and Artificial Intelligence. *Cancers* **2021**, *13*, 3162. [CrossRef]

134. Zhang, S.M.; Wang, Y.J.; Zhang, S.T. Accuracy of artificial intelligence-assisted detection of esophageal cancer and neoplasms on endoscopic images: A systematic review and meta-analysis. *J. Dig. Dis.* **2021**, *22*, 318–328. [CrossRef]

135. Guleria, S.; Shah, T.U.; Pulido, J.V.; Fasullo, M.; Ehsan, L.; Lippman, R.; Sali, R.; Mutha, P.; Cheng, L.; Brown, D.E.; et al. Deep learning systems detect dysplasia with human-like accuracy using histopathology and probe-based confocal laser endomicroscopy. *Sci. Rep.* **2021**, *11*, 5086. [CrossRef]

136. Yu, H.; Singh, R.; Shin, S.H.; Ho, K.Y. Artificial intelligence in upper GI endoscopy—Current status, challenges and future promise. *J. Gastroenterol. Hepatol.* **2021**, *36*, 20–24. [CrossRef] [PubMed]

137. Arribas, J.; Antonelli, G.; Frazzoni, L.; Fuccio, L.; Ebigbo, A.; van der Sommen, F.; Ghatwary, N.; Palm, C.; Coimbra, M.; Renna, F.; et al. Standalone performance of artificial intelligence for upper GI neoplasia: A meta-analysis. *Gut* **2020**, *70*, 1458–1468. [CrossRef]

138. Wu, L.; He, X.; Liu, M.; Xie, H.; An, P.; Zhang, J.; Zhang, H.; Ai, Y.; Tong, Q.; Guo, M.; et al. Evaluation of the effects of an artificial intelligence system on endoscopy quality and preliminary testing of its performance in detecting early gastric cancer: A randomized controlled trial. *Endoscopy* **2021**. [CrossRef] [PubMed]

139. Tang, D.; Wang, L.; Ling, T.; Lv, Y.; Ni, M.; Zhan, Q.; Fu, Y.; Zhuang, D.; Guo, H.; Dou, X.; et al. Development and validation of a real-time artificial intelligence-assisted system for detecting early gastric cancer: A multicentre retrospective diagnostic study. *EBioMedicine* **2020**, *62*, 103146. [CrossRef] [PubMed]

140. Ali, S.; Bailey, A.; Ash, S.; Haghighat, M.; Leedham, S.J.; Lu, X.; East, J.E.; Rittscher, J.; Braden, B. A Pilot Study on Automatic Three-Dimensional Quantification of Barrett's Esophagus for Risk Stratification and Therapy Monitoring. *Gastroenterology* **2021**, *161*, 865–878.e8. [CrossRef]

141. Waterhouse, D.J.; Januszewicz, W.; Ali, S.; Fitzgerald, R.C.; di Pietro, M.; Bohndiek, S.E. Spectral Endoscopy Enhances Contrast for Neoplasia in Surveillance of Barrett's Esophagus. *Cancer Res.* **2021**, *81*, 3415–3425. [CrossRef]

142. Tang, D.; Zhou, J.; Wang, L.; Ni, M.; Chen, M.; Hassan, S.; Luo, R.; Chen, X.; He, X.; Zhang, L.; et al. A Novel Model Based on Deep Convolutional Neural Network Improves Diagnostic Accuracy of Intramucosal Gastric Cancer (with Video). *Front. Oncol.* **2021**, *11*, 622827. [CrossRef]

143. Zhou, C.; Hu, J.; Wang, Y.; Ji, M.H.; Tong, J.; Yang, J.J.; Xia, H. A machine learning-based predictor for the identification of the recurrence of patients with gastric cancer after operation. *Sci. Rep.* **2021**, *11*, 1571. [CrossRef]

144. Bang, C.S.; Lee, J.J.; Baik, G.H. Artificial Intelligence for the Prediction of Helicobacter Pylori Infection in Endoscopic Images: Systematic Review and Meta-Analysis Of Diagnostic Test Accuracy. *J. Med. Internet Res.* **2020**, *22*, e21983. [CrossRef]

145. Sundar, R.; Barr Kumarakulasinghe, N.; Huak Chan, Y.; Yoshida, K.; Yoshikawa, T.; Miyagi, Y.; Rino, Y.; Masuda, M.; Guan, J.; Sakamoto, J.; et al. Machine-learning model derived gene signature predictive of paclitaxel survival benefit in gastric cancer: Results from the randomised phase III SAMIT trial. *Gut* **2021**. [CrossRef]

146. Chen, H.; Luo, J.; Guo, J. Construction and Validation of a 7-Immune Gene Model for Prognostic Assessment of Esophageal Carcinoma. *Med. Sci. Monit.* **2020**, *26*, e927392. [CrossRef] [PubMed]

147. Liu, S.; Wu, J.; Xia, Q.; Liu, H.; Li, W.; Xia, X.; Wang, J. Finding new cancer epigenetic and genetic biomarkers from cell-free DNA by combining SALP-seq and machine learning. *Comput. Struct. Biotechnol. J.* **2020**, *18*, 1891–1903. [CrossRef] [PubMed]

148. Li, Z.; Gao, X.; Peng, X.; May Chen, M.J.; Li, Z.; Wei, B.; Wen, X.; Wei, B.; Dong, Y.; Bu, Z.; et al. Multi-omics characterization of molecular features of gastric cancer correlated with response to neoadjuvant chemotherapy. *Sci. Adv.* **2020**, *6*, eaay4211. [CrossRef] [PubMed]

149. Yuan, L.; Zhao, J.; Sun, T.; Shen, Z. A machine learning framework that integrates multi-omics data predicts cancer-related LncRNAs. *BMC Bioinform.* **2021**, *22*, 332. [CrossRef]

150. Li, Q.; Liu, X.; Gu, J.; Zhu, J.; Wei, Z.; Huang, H. Screening lncRNAs with diagnostic and prognostic value for human stomach adenocarcinoma based on machine learning and mRNA-lncRNA co-expression network analysis. *Mol. Genet. Genom. Med.* **2020**, *8*, e1512. [CrossRef]

151. Yaoxing, H.; Danchun, Y.; Xiaojuan, S.; Shuman, J.; Qingqing, Y.; Lin, J. Identification of Novel Susceptible Genes of Gastric Cancer Based on Integrated Omics Data. *Front. Cell Dev. Biol.* **2021**, *9*, 712020. [CrossRef] [PubMed]

152. Wang, H.; Ding, Y.; Chen, Y.; Jiang, J.; Chen, Y.; Lu, J.; Kong, M.; Mo, F.; Huang, Y.; Zhao, W.; et al. A novel genomic classification system of gastric cancer via integrating multidimensional genomic characteristics. *Gastric Cancer* **2021**. [CrossRef]

153. Zhang, G.; Xue, Z.; Yan, C.; Wang, J.; Luo, H. A Novel Biomarker Identification Approach for Gastric Cancer Using Gene Expression and DNA Methylation Dataset. *Front. Genet.* **2021**, *12*, 644378. [CrossRef]

154. Owen, R.P.; White, M.J.; Severson, D.T.; Braden, B.; Bailey, A.; Goldin, R.; Wang, L.M.; Ruiz-Puig, C.; Maynard, N.D.; Green, A.; et al. Single cell RNA-seq reveals profound transcriptional similarity between Barrett's oesophagus and oesophageal submucosal glands. *Nat. Commun.* **2018**, *9*, 4261. [CrossRef]

155. Kiselev, V.Y.; Kirschner, K.; Schaub, M.T.; Andrews, T.; Yiu, A.; Chandra, T.; Natarajan, K.N.; Reik, W.; Barahona, M.; Green, A.R.; et al. SC3: Consensus clustering of single-cell RNA-seq data. *Nat. Methods* **2017**, *14*, 483–486. [CrossRef]

156. Chen, Y.; Wei, K.; Liu, D.; Xiang, J.; Wang, G.; Meng, X.; Peng, J. A Machine Learning Model for Predicting a Major Response to Neoadjuvant Chemotherapy in Advanced Gastric Cancer. *Front. Oncol.* **2021**, *11*, 675458. [CrossRef]

157. Jiang, Y.; Liang, X.; Han, Z.; Wang, W.; Xi, S.; Li, T.; Chen, C.; Yuan, Q.; Li, N.; Yu, J.; et al. Radiographical assessment of tumour stroma and treatment outcomes using deep learning: A retrospective, multicohort study. *Lancet Digit. Health* **2021**, *3*, e371–e382. [CrossRef]

158. Xu, Q.; Sun, Z.; Li, X.; Ye, C.; Zhou, C.; Zhang, L.; Lu, G. Advanced gastric cancer: CT radiomics prediction and early detection of downstaging with neoadjuvant chemotherapy. *Eur. Radiol.* **2021**. [CrossRef] [PubMed]

159. Tan, J.W.; Wang, L.; Chen, Y.; Xi, W.; Ji, J.; Wang, L.; Xu, X.; Zou, L.K.; Feng, J.X.; Zhang, J.; et al. Predicting Chemotherapeutic Response for Far-advanced Gastric Cancer by Radiomics with Deep Learning Semi-automatic Segmentation. *J. Cancer* **2020**, *11*, 7224–7236. [CrossRef] [PubMed]

160. Takeuchi, M.; Seto, T.; Hashimoto, M.; Ichihara, N.; Morimoto, Y.; Kawakubo, H.; Suzuki, T.; Jinzaki, M.; Kitagawa, Y.; Miyata, H.; et al. Performance of a deep learning-based identification system for esophageal cancer from CT images. *Esophagus* **2021**, *18*, 612–620. [CrossRef]

161. Liu, S.; Qiao, X.; Xu, M.; Ji, C.; Li, L.; Zhou, Z. Development and Validation of Multivariate Models Integrating Preoperative Clinicopathological Parameters and Radiographic Findings Based on Late Arterial Phase CT Images for Predicting Lymph Node Metastasis in Gastric Cancer. *Acad. Radiol.* **2021**. [CrossRef]

162. Mirniaharikandehei, S.; Heidari, M.; Danala, G.; Lakshmivarahan, S.; Zheng, B. Applying a random projection algorithm to optimize machine learning model for predicting peritoneal metastasis in gastric cancer patients using CT images. *Comput. Methods Programs Biomed.* **2021**, *200*, 105937. [CrossRef] [PubMed]

163. Huang, Z.; Liu, D.; Chen, X.; He, D.; Yu, P.; Liu, B.; Wu, B.; Hu, J.; Song, B. Deep Convolutional Neural Network Based on Computed Tomography Images for the Preoperative Diagnosis of Occult Peritoneal Metastasis in Advanced Gastric Cancer. *Front. Oncol.* **2020**, *10*, 601869. [CrossRef] [PubMed]

164. Sun, R.J.; Fang, M.J.; Tang, L.; Li, X.T.; Lu, Q.Y.; Dong, D.; Tian, J.; Sun, Y.S. CT-based deep learning radiomics analysis for evaluation of serosa invasion in advanced gastric cancer. *Eur. J. Radiol.* **2020**, *132*, 109277. [CrossRef] [PubMed]

165. Jang, H.-J.; Song, I.-H.; Lee, S.-H. Deep Learning for Automatic Subclassification of Gastric Carcinoma Using Whole-Slide Histopathology Images. *Cancers* **2021**, *13*, 3811. [CrossRef]

166. Sharma, H.; Zerbe, N.; Klempert, I.; Hellwich, O.; Hufnagl, P. Deep convolutional neural networks for automatic classification of gastric carcinoma using whole slide images in digital histopathology. *Comput. Med. Imaging Graph.* **2017**, *61*, 2–13. [CrossRef]

167. Wang, X.; Chen, Y.; Gao, Y.; Zhang, H.; Guan, Z.; Dong, Z.; Zheng, Y.; Jiang, J.; Yang, H.; Wang, L.; et al. Predicting gastric cancer outcome from resected lymph node histopathology images using deep learning. *Nat. Commun.* **2021**, *12*, 1637. [CrossRef] [PubMed]

168. Klein, S.; Gildenblat, J.; Ihle, M.A.; Merkelbach-Bruse, S.; Noh, K.W.; Peifer, M.; Quaas, A.; Büttner, R. Deep learning for sensitive detection of Helicobacter Pylori in gastric biopsies. *BMC Gastroenterol.* **2020**, *20*, 417. [CrossRef]

169. Park, J.; Jang, B.G.; Kim, Y.W.; Park, H.; Kim, B.H.; Kim, M.J.; Ko, H.; Gwak, J.M.; Lee, E.J.; Chung, Y.R.; et al. A Prospective Validation and Observer Performance Study of a Deep Learning Algorithm for Pathologic Diagnosis of Gastric Tumors in Endoscopic Biopsies. *Clin. Cancer Res.* **2021**, *27*, 719–728. [CrossRef]

170. Song, Z.; Zou, S.; Zhou, W.; Huang, Y.; Shao, L.; Yuan, J.; Gou, X.; Jin, W.; Wang, Z.; Chen, X.; et al. Clinically applicable histopathological diagnosis system for gastric cancer detection using deep learning. *Nat. Commun.* **2020**, *11*, 4294. [CrossRef] [PubMed]

171. Sali, R.; Moradinasab, N.; Guleria, S.; Ehsan, L.; Fernandes, P.; Shah, T.U.; Syed, S.; Brown, D.E. Deep Learning for Whole-Slide Tissue Histopathology Classification: A Comparative Study in the Identification of Dysplastic and Non-Dysplastic Barrett's Esophagus. *J. Pers. Med.* **2020**, *10*, 141. [CrossRef]

172. Meier, A.; Nekolla, K.; Hewitt, L.C.; Earle, S.; Yoshikawa, T.; Oshima, T.; Miyagi, Y.; Huss, R.; Schmidt, G.; Grabsch, H.I. Hypothesis-free deep survival learning applied to the tumour microenvironment in gastric cancer. *J. Pathol. Clin. Res.* **2020**, *6*, 273–282. [CrossRef]

173. Kather, J.N.; Calderaro, J. Development of AI-based pathology biomarkers in gastrointestinal and liver cancer. *Nat. Rev. Gastroenterol. Hepatol.* **2020**, *17*, 591–592. [CrossRef]

174. Calderaro, J.; Kather, J.N. Artificial intelligence-based pathology for gastrointestinal and hepatobiliary cancers. *Gut* **2021**, *70*, 1183–1193. [CrossRef] [PubMed]

175. Kuntz, S.; Krieghoff-Henning, E.; Kather, J.N.; Jutzi, T.; Höhn, J.; Kiehl, L.; Hekler, A.; Alwers, E.; von Kalle, C.; Fröhling, S.; et al. Gastrointestinal cancer classification and prognostication from histology using deep learning: Systematic review. *Eur. J. Cancer* **2021**, *155*, 200–215. [CrossRef]

176. Muti, H.S.; Heij, L.R.; Keller, G.; Kohlruss, M.; Langer, R.; Dislich, B.; Cheong, J.-H.; Kim, Y.-W.; Kim, H.; Kook, M.-C.; et al. Development and validation of deep learning classifiers to detect Epstein-Barr virus and microsatellite instability status in gastric cancer: A retrospective multicentre cohort study. *Lancet Digit. Health* **2021**, *3*, e654–e664. [CrossRef]

177. Kather, J.N.; Pearson, A.T.; Halama, N.; Jäger, D.; Krause, J.; Loosen, S.H.; Marx, A.; Boor, P.; Tacke, F.; Neumann, U.P.; et al. Deep learning can predict microsatellite instability directly from histology in gastrointestinal cancer. *Nat. Med.* **2019**, *25*, 1054–1056. [CrossRef]

178. Patel, S.K.; George, B.; Rai, V. Artificial Intelligence to Decode Cancer Mechanism: Beyond Patient Stratification for Precision Oncology. *Front. Pharm.* **2020**, *11*, 1177. [CrossRef]

179. Zeng, D.; Li, M.; Zhou, R.; Zhang, J.; Sun, H.; Shi, M.; Bin, J.; Liao, Y.; Rao, J.; Liao, W. Tumor Microenvironment Characterization in Gastric Cancer Identifies Prognostic and Immunotherapeutically Relevant Gene Signatures. *Cancer Immunol. Res.* **2019**, *7*, 737–750. [CrossRef]

180. Shi, X.-J.; Wei, Y.; Ji, B. Systems Biology of Gastric Cancer: Perspectives on the Omics-Based Diagnosis and Treatment. *Front. Mol. Biosci.* **2020**, *7*, 203. [CrossRef] [PubMed]

181. Abadjian, M.Z.; Edwards, W.B.; Anderson, C.J. Imaging the Tumor Microenvironment. *Adv. Exp. Med. Biol.* **2017**, *1036*, 229–257. [CrossRef]

182. Zhou, Z.; Lu, Z.-R. Molecular imaging of the tumor microenvironment. *Adv. Drug Deliv. Rev.* **2017**, *113*, 24–48. [CrossRef]

183. Ramamonjisoa, N.; Ackerstaff, E. Characterization of the Tumor Microenvironment and Tumor-Stroma Interaction by Non-invasive Preclinical Imaging. *Front. Oncol.* **2017**, *7*, 3. [CrossRef]

184. Han, J.; Zhao, Y.; Shirai, K.; Molodtsov, A.; Kolling, F.W.; Fisher, J.L.; Zhang, P.; Yan, S.; Searles, T.G.; Bader, J.M.; et al. Resident and circulating memory T cells persist for years in melanoma patients with durable responses to immunotherapy. *Nat. Cancer* **2021**, *2*, 300–311. [CrossRef]

185. Jain, R.K.; Duda, D.G.; Willett, C.G.; Sahani, D.V.; Zhu, A.X.; Loeffler, J.S.; Batchelor, T.T.; Sorensen, A.G. Biomarkers of response and resistance to antiangiogenic therapy. *Nat. Rev. Clin. Oncol.* **2009**, *6*, 327–338. [CrossRef] [PubMed]

186. Cleary, J.M.; Horick, N.K.; McCleary, N.J.; Abrams, T.A.; Yurgelun, M.B.; Azzoli, C.G.; Rubinson, D.A.; Brooks, G.A.; Chan, J.A.; Blaszkowsky, L.S.; et al. FOLFOX plus ziv-aflibercept or placebo in first-line metastatic esophagogastric adenocarcinoma: A double-blind, randomized, multicenter phase 2 trial. *Cancer* **2019**, *125*, 2213–2221. [CrossRef] [PubMed]

187. Gerke, S.; Babic, B.; Evgeniou, T.; Cohen, I.G. The need for a system view to regulate artificial intelligence/machine learning-based software as medical device. *NPJ Digit. Med.* **2020**, *3*, 53. [CrossRef] [PubMed]

188. Selvaraju, R.R.; Cogswell, M.; Das, A.; Vedantam, R.; Parikh, D.; Batra, D. Grad-CAM: Visual Explanations from Deep Networks via Gradient-Based Localization. In Proceedings of the 2017 IEEE International Conference on Computer Vision (ICCV), Venice, Italy, 22–29 October 2017; pp. 618–626.

189. Rudin, C. Stop explaining black box machine learning models for high stakes decisions and use interpretable models instead. *Nat. Mach. Intell.* **2019**, *1*, 206–215. [CrossRef]

190. Klein, S.; Quaas, A.; Quantius, J.; Löser, H.; Meinel, J.; Peifer, M.; Wagner, S.; Gattenlöhner, S.; Wittekindt, C.; von Knebel Doeberitz, M.; et al. Deep Learning Predicts HPV Association in Oropharyngeal Squamous Cell Carcinomas and Identifies Patients with a Favorable Prognosis Using Regular H&E Stains. *Clin. Cancer Res.* **2021**, *27*, 1131–1138. [CrossRef]

191. London, A.J. Artificial Intelligence and Black-Box Medical Decisions: Accuracy versus Explainability. *Hastings Cent. Rep.* **2019**, *49*, 15–21. [CrossRef]

192. Ramesh, A.N.; Kambhampati, C.; Monson, J.R.; Drew, P.J. Artificial intelligence in medicine. *Ann. R. Coll. Surg. Engl.* **2004**, *86*, 334–338. [CrossRef]

193. Elhassan, A.; Abu-Soud, S.M.; Alghanim, F.; Salameh, W. ILA4: Overcoming missing values in machine learning datasets—An inductive learning approach. *J. King Saud. Univ.-Comput. Inf. Sci.* **2021**. [CrossRef]

194. Goh, W.W.B.; Wang, W.; Wong, L. Why Batch Effects Matter in Omics Data, and How to Avoid Them. *Trends Biotechnol.* **2017**, *35*, 498–507. [CrossRef] [PubMed]