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Poorly cohesive cells gastric carcinoma including signet-ring cell cancer: updated review of definition, classification and therapeutic management.

poorly cohesive cells gastric carcinoma review

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Abstract

While the incidence of gastric cancer (GC) in general has decreased worldwide in recent decades, the incidence of diffuse cancer historically comprising poorly cohesive cells gastric cancer (PCC-GC) and including signet ring cell cancer is rising. Literature concerning PCC-GC is scarce and unclear, mostly due to a large variety of historically used definitions and classifications. Compared to other histological subtypes of GC, PCC-GC is nevertheless characterized by a distinct set of epidemiological, histological and clinical features, which require a specific diagnostic and therapeutic approach. The aim of this review was to provide an update on the definition, classification and therapeutic strategies of PCC-GC. We focus on the updated histological definition of PCC-GC, along with its implications on future treatment strategies and study design. Also, specific considerations in the diagnostic management are discussed. Finally, the impact of some recent developments in the therapeutic management of GC in general such as the recently validated taxane-based regimens (FLOT), the use of hyperthermic intraperitoneal chemotherapy (HIPEC) as well as Pressurized IntraPeritoneal Aerosol Chemotherapy (PIPAC) and targeted therapy have been reviewed in depth for their relative importance for PCC-GC in particular.

Key Words: poorly cohesive cells gastric carcinoma; review; definition; classification; therapeutic management

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Core Tip: Although the worldwide incidence of gastric cancer has decreased in recent decades, the incidence of diffuse cancer historically comprising Poorly Cohesive Cells Gastric Cancer (PCC-GC) and including Signet Ring Cell cancer is rising. While the existing literature concerning PCC-GC is scarce, this narrative review aims to provide

an update on the classification and management of PCC-GC in light of several recent developments: (i) the updated definition according to World Health Organization classification and Verona consensus, (ii) an update in curative approaches following recent validation of FLOT regimen and development of hyperthermic intraperitoneal chemotherapy (HIPEC), and (iii) role of chemotherapy and targeted therapies in the treatment of PCC-GC.

INTRODUCTION

Introduction

Worldwide, gastric cancer (GC) is ranked as the 5th most frequently diagnosed cancer. Because of its poor prognosis, it is responsible for the 3nd highest cancer related death rate [1]. Despite a global decline in the overall incidence of GC, the relative incidence of diffuse type GC historically comprising Poorly Cohesive Cells GC (PCC-GC) and including Signet Ring Cell (SRC) cancer has shown a steady increase in the past few decades, especially in the United States and Europe [2–4]. Based on data from the Surveillance, Epidemiology, and End Results (SEER) database, collected between 1973 and 2000, an increase of 400% of the diffuse type GC has been noted [4]. In contrast to other histological types of GC, SRC-GC is known to be associated with a younger age at the time of diagnosis along with a more female gender distribution [5–8]. Since the publication of the first edition of the World Health Organization (WHO) classification of GC in 1977, the definition of SRC-GC has changed several times until the 5th edition in 2019 [9–13]. Before 2010, SRC-GC was classified as a separate specific subtype of GC [9,10,13]. In the edition of 2010, the SRC-GC category was completely redefined as a subtype of PCC-GC [10]. Previously, alternative classification systems, such as the Lauren and the Ming classification, categorized SRC-GC as 'diffuse/mixed' and 'infiltrative' type carcinoma, respectively [14,15]. As such, these multiple definitions and classifications render correct assessment and comparison of this histological subtype in the current literature difficult to make. In this context, an updated review on PCC-GC was needed to addres the following topics: (i) recent definition according to WHO

classification [12] and Verona consensus [16], (ii) update in curative approaches following validation of the new perioperative chemotherapy (CT) regimen 5-Fluorouracil, Leucovorin, Oxaliplatin and Docetaxel (FLOT) [17,18] and the increasing role of Hiperthermic IntraPeritoneal Chemotherapy (HIPEC) in the prevention of or as a curative treatment for peritoneal metastases, and (iii) recent developments in future-based therapeutic strategies including chemotherapy, Pressurized IntraPeritoneal Aerosol Chemotherapy (PIPAC) and targeted therapies including immunotherapy.

Methods

A literature search in the MEDLINE/PubMed database was conducted with the use of the following search terms: 'signet ring cell carcinoma' (n = 3345), 'poorly cohesive cells' (n = 136), 'Lauren and diffuse type' (n = 257), 'linitis plastica' (n = 423) and 'Bormann type IV' (n = 178) up to 2021 . Only studies in English language, published after January 1980 were eligible for inclusion. Studies were screened based on the abstract. Additional studies were retrieved by screening the references of each individual article. Case reports and studies including patients <18 years old were excluded as well as studies reporting on non-gastric PCC-GC. Studies reporting on < 30 cases were also excluded. Abstracts and meeting reports were only included if the information was found to be relevant enough in the context of the subject. Studies were only included after agreement of both VD and GP.

Results

Overview and update on histological and molecular classifications of SRC- and PCC-GC.

The most commonly used classifications in GC are the WHO and the Laurén classifications [10,14][10,11].

WHO and Verona classifications

The WHO definition of SRC-GC and - more recently - PCC-GC has evolved over time in function of the different published editions of the WHO classification. In the very first edition, published in 1977, SRC-GC was considered as a separate subtype of GC and was defined as 'a tumor which contained more than 50% of isolated or small groups of

malignant cells containing intracytoplasmic mucin'. As such, 4 morphological SRC types were defined [9]. By the time the 3rd edition of the WHO classification was published in 2000, this was extended to 5 morphological SRC types [11]. In the 4th edition in 2010, the SRC-GC category was completely redefined as a subtype of PCC-GC [10,12]. PCC-GC are composed of neoplastic cells that are isolated or arranged in small aggregates without well-formed glands. The definition of the extent of SRC to qualify as SRC-GC became "predominantly" or "exclusively" in the 4th and 5th editions of the WHO [10,12]. SRCs are characterized by a central optically clear, globoid droplet of cytoplasmic mucin with an eccentrically placed nucleus [10]. Other cellular subtypes not fulfilling the requirements of this definition, should be defined as PCC Not Otherwise Specified (PCC-NOS). PCC-NOS include tumors composed of neoplastic cells resembling histiocytes or lymphocytes; others have deeply esosinophilic cytoplasm; some PCC are pleomorphic with bizarre nuclei. A mixture of the different cell types can be seen, including a mixture of PCC-NOS and SRC. Historically, mucinous adenocarcinoma has frequently been misclassified as SRCC, due to the frequent observation of SRC into this subtype [19,20]. Overall this added a lot confusion in analyzing data from the literature.

Invited by the European chapter of the International Gastric Cancer Association (IGCA), a multidisciplinary expert panel convened in 2017 with the intent to clarify the pathological definition of PCC-GC [16]. In a consented final conclusion, it was proposed that only PCC-GC with more than 90% of cells representing a SRC morphology, should be classified as SRC-type [16]. The two other categories were PCC with SRC component (<90% but >10% of SRC) and PCC-NOS: <10% of SRC. An overview of the proposed definition and classification is shown in table 1 and figure 1. On another level, this newly defined classification also incorporates the theory that the extent of SRC in the tumour may be an expression of the differentiation grade of PCC [16]. The importance of this consensus definition cannot be underestimated, since it will enable future studies to standardize results and facilitate comparison between studies in order to avoid the

major heterogeneity that has characterized studies concerning SRC-GC for the past few decades.

Laurén and other classifications

The Laurén classification, which is the oldest and most widespread classification, categorizes tumours into two major categories: intestinal type tumors, characterized by cohesive neoplastic cells organized in well-differentiated glandular structures and diffuse tumors, diffusely infiltrating the gastric wall, with little to no gland formation. The latter type consists of PCCs, with or without SRC morphology and thus corresponds the most with the PCC category of the WHO classification [14]. Comparative studies are showed in Table 2. Tumors exhibiting features of both the intestinal and diffuse types (>25% of either component) are designated as mixed-type adenocarcinoma and account for approximately 10% of all gastric adenocarcinomas [21,22]. Some tumors may be unclassified. Although widely implemented, the Laurén classification does not allow for any clinical or pathological evaluation according to the proportion of SRC component, which is an additional justification for the implantation of the recently proposed renewed definition of PCC by the WHO [12] and the European chapter of IGCA [16].

The original Japanese classification system categorized GC into differentiated and undifferentiated tumors with undifferentiated type corresponding to diffuse type [23]. A more recent version of the classification proposed by the Japanese Gastric Cancer Association (JCGA) is however mainly based on the WHO classification and distinguishes between papillary, tubular, poorly differentiated and mucinous adenocarcinoma as well as SRC tumors [24]. Finally the Ming classification describing expanding and infiltrative type, the latter being strongly correlated to diffuse type [25,26].

Linitis plastica

Linitis plastica (LP) is macroscopically described as an increase thickening and rigidity of the gastric walls with an aspect of linen. From a histological point of view, it corresponds to an involvement of the entire stomach wall by carcinoma cells, mostly SRC, with a very abundant sclerous stroma. LP is an uncommon variant of GA

occurring in 7–17.4 % of cases [8,27–40]. LP is rarely individualised in studies for two main reasons; (i) some authors confuse the histological and macroscopical definition [41–43] assimilating SRC-GC with LP, thus adding to the confusion and (ii) LP is also referred to as Borrmann type IV or scirrhous gastric carcinoma in the Eastern litterature. An illustration of gastric LP is presented in figure 2. In one study at our center, among 159 patients with SRC-GC and non-SRC_GC, LP occurred in 35.6% in the SRC group *vs* 6% in the non SRCC group ($p<0.001$) [44]. Most of LP in the non SRC-group had a minor component of SRC. In other words, LP and SRCC are not synonyms [45] but are however closely associated. However, we believe that the current definition of SCR-GC should be used systematically. The term 'linitis plastica' can be additionaly used when applicable.

Molecular characteristics

From a molecular point of view, GG has been classified into four genomic subtypes in a landmark project by The Cancer Genome Atlas (TCGA) [46]. These four subtypes comprise: (i) the Epstein-Barr (EBV) subtype (9%), characterized by extreme DNA hypermethylation, recurrent PIK3CA mutations and amplification of JAK2, PD-L1 and PD-L2; (ii) the microsatellite instability (MSI) subtype (21%), containing mutations in genes encoding for targetable oncogenic signalling proteins and associated with a more favorable oncological outcome; (iii) a genomically stable (GS) subtype (20%), in which most but not all PCC-GC are categorized and (iv) the chromosomal instability (CIN) subtype (50%), associated with aneuploidy and amplifications of genes involved in receptor tyrosine kinase/RAS/MAPK signalling [47]. More recently, another molecular analysis for GC identified four subgroups of tumors associated with distinct clinical outcomes: (i) a mesenchymal-type, including diffuse-subtype tumors and most PCC-GC tumors; (ii) a MSI subtype, characterized by numerous mutations and a better prognosis; (iii) a tumor protein 53 (TP53)-active subtype, associated with higher rates of EBV infection and (iv) a TP53-inactive subtype, similar to the CIN subgroup [48]. The importance of these molecular classifications cannot be underestimated as they provide

a roadmap for patient stratification. In addition to the prognostic impact, it has been proven that these genomic subtypes are associated with distinct features regarding tumor response. As such, this subtype classification is primordial in the implementation of current and future clinical trials that evaluate the role of targeted therapies amongst others [49,50]. However we have to bear in mind that GC consists of heterogenous tumours and that several histological and molecular components can be present in the same tumor and may be modified by the treatment applied. [51] In addition, there is no strict correlation between histological types and molecular subtypes. PCC-GC are mostly GS but can also be MSI or EBV type with potential therapeutic implications since both molecular subtypes are associated with response to immune checkpoint inhibitors [52].

Prognostic features of PCC-GC

All stages studies

Although most studies agree about the poor prognosis of diffuse GC according to the Laurén classification, more discrepancies exist about the specific prognosis of PCC-GC [22,44,53,54]. An overview of studies reporting on the prognosis of all stages SRC- and PCC-GC, is shown in table 3. The reported prognosis of PCC-GC in Western studies is in general worse compared to that of most Eastern studies with however significant differences in terms of tumor stages; the majority of studies in early gastric cancer (EGC) (i.e. GC pT1a or pT1b regardless of lymph node status) [55] originate from Eastern series.

Among PCC tumors, the prognostic impact of the relative percentages of an SRC component within the tumors remains controversial [49]. Two studies evaluated the prognostic role of the Verona consensus with marked differences between the distribution of the 3 categories questioning reproducibility of the classification (Table 1 [49,56,57]. Bencivenga showed that the percentage of SRC was associated with tumour stage and survival in PCC-GC: the percentage of SRC was inversely related to tumour aggressiveness, pT stage ($P < .001$) and the number of positive nodes coded as a continuous variable ($P = 0.009$). Long term survival was significantly higher in SRC-

type (>90% SRC) compared with PCC with SRC component (<90% but >10% of SRC) and PCC-NOS (<10% of SRC) tumors [56]. In the other study, pathological revision found no patients with SRC-type (>90% SRC) [57]. Five-years overall survival was significantly higher in PCC with SRC component (<90% but >10% of SRC) compared with PCC-NOS (<10% of SRC) (63.3% vs. 12.7%) [57].

Early Gastric Cancer

An overview of studies reporting on the prognostic outcomes of SRC- or PCC-EGC is shown in table 4. Most studies demonstrated that the prognosis of SRC- or PCC-EGC is similar or even better than that of other EGC [58–61]. The largest of these studies, including data on 3272 patients, concluded that the prognosis of SRC-EGC was better than that of well-and moderately-differentiated EGC (HR for overall survival (OS)=0.66, 95%CI [0.44-0.98]) [62]. In one of the few Western studies, Gronnier *et al* showed that SRC-EGC was associated with a 5y-OS benefit (85% vs. 76%, P = 0.035) compared to non-SRCEGC, although SRC-EGC was more frequently associated with submucosal invasion [63]. The survival benefit in this study was however no longer objectivated after multivariable analysis, possibly because of the lower rate of non-cancer related deaths in the younger SRC group. More studies in Western populations are required to further validate the superior prognostic results of PCC- or SRC-EGC as reported by Eastern series and should include an analysis according to the new WHO classification and Verona consensus [12,16].

Advanced gastric cancer (GC invading beyond the submucosa)

Table 5 presents an overview of studies reporting on the prognostic characteristics of SRC- or PCC-AGC. At and advanced stage, SRC-AGC is associated with deeper tumor invasion, a higher rate of lymph node involvement, an increased potential for diffuse infiltration of the gastric wall (LP), a greater risk of peritoneal metastatic disease, lower rates of R0 resection and higher rates of early disease recurrence [53,64–66]. Whether the dismal prognosis of PCC-GC is related to a more advanced stage of the disease at the time of diagnosis or to an inherently more aggressive tumor biology, is much debated [44,53]. Results from a large population-based study in the United States demonstrated

that, after adjustment for stage, SRC histology was not independently associated with worse prognosis [53]. These findings seem to be confirmed by several other studies that reported a worse prognosis in a univariable analysis, but not in a multivariable analysis after adjustment for tumor stage [6,65–67]. Critics however state that a posteriori adjustment by multivariable analysis results in an oversimplification of the issue. In the absence of any possibility for prospective randomization, some authors stated that a matched case-control analysis should be the methodological tool of choice to clarify this debate [68]. As such, Piessen *et al* confirmed that SRC histology entailed a worse stage-independent prognosis in patients with GC compared to other histological subtypes [44].

The underlying factors that may cause the discrepancy between the prognostic characteristics of early and advanced PCC-GC remain uncertain. This topic is even more complicated by the geographical differences and potential variability in the molecular tumor characteristics between Western and Eastern populations [69]. Within the group of GC, early and advanced PCC-GC may represent two distinct entities, each with its own prognostic features [70].

Pre-therapeutic evaluation in PCC-GC

Thorough anamnestic evaluation with emphasis on family history should be performed in order to detect clinical criteria for hereditary diffuse gastric cancer [71]. Because the tumoral spread in PCC-GC mostly occurs within the deeper tissue layers, mostly in the absence of any mucosal alterations, conventional endoscopy and superficial biopsies may miss the diagnosis. Repeated endoscopies should consequently be performed along with deep biopsies, guided by endoscopic ultrasonography. A CT scan can give useful additional information by identifying areas of the stomach characterized by an increased wall thickness in case of LP.

In light of the WHO criteria from 2000 for SRC-GC (i.e more than 50% SRC), the overall reliability of pretherapeutic biopsies to predict specimen histology has been evaluated. Among 254 patients, presence of SRC in routine pre-therapeutic endoscopic biopsies could accurately predict SRC histology and its associated poor prognosis (Sensitivity: 88.1%, Specificity: 95.4%, Positive predictive value: 92.7%, Negative predictive value:

92.4%) [5]. Future studies evaluating the concordance between pretherapeutic biopsies and specimen in PCC-GC will have to be performed using the new WHO definition and the Verona consensus [12,16].

Positron emission tomography (PET) imaging using fluoro-2-deoxy-D-glucose (FDG) may be useful to eliminate distant metastases in case of advanced disease [72,73]. However, PCC-GC has proven to be associated with a lower PET-sensitivity and a lower standard uptake value (SUV) than no PCC-GC with consequently a potential risk of false negative results. [74–76]. In addition two studies suggested that a higher SUVmax was a predictive factor of poor prognosis in SRC histology [77,78].

Staging laparoscopy is currently recommended by the European society for Medical Oncology (ESMO) for tumors ≥ stage Ib [79] and by the National Comprehensive Cancer Network (NCCN) for tumors ≥ T1b [80]. Several studies reported high rates of peritoneal carcinomatosis (5-21%) discovered during surgical exploration after a standard work-up including CT-scan in advanced PCC-GC or diffuse tumors [44,81–83] In the Plastic study, comparing staging laparoscopy and FDG-PET/CT in preoperative workup of locally advanced gastric cancer, treatment intent changed from curative to palliative in 73 patients (19%) after staging laparoscopy (detecting peritoneal or locally non-resectable disease) *vs* in 12 patients (3%) FDG-PET/CT (detecting distant metastases) [83]. This risk was 1.5 to 3 times higher than in other tumors [44,83]. Staging laparoscopy has been consequently proposed as an important tool for pretherapeutic evaluation of PCC-GC [84]. In addition to a complete and systematic exploration of the abdominal cavity, staging laparoscopy provides the possibility to perform a peritoneal lavage with cytology. A positive cytology classifies the disease as stage IV, necessitating a change in therapeutic strategy [31,39,85]. Alternative procedures such as laparo-endoscopic single site surgery are currently being evaluated in order to optimize the detection of peritoneal disease even further, since even with standard staging laparoscopy, lesions on the mesenteric side of the small bowel are still frequently missed [86,87]. A small periumbilical incision to explore the small bowel by means of palpation may be helpful in advanced PCC-GC.

Curative treatment

Endoscopic resection

An increasing amount of evidence has been gathered that endoscopic treatment by means of an endoscopic submucosal dissection (ESD) could represent a valid option for non-ulcerated undifferentiated lesions, ≤ 2 cm in diameter, limited to the mucosa and without LVI [59,88–90]. Lesions in this category are currently excluded from the absolute indication by the Japanese Gastric Cancer Association (JGCA) recommendations due to the lack of sufficient evidence for long-term outcome, but may in future be included pending the results of the JCOG1009/1010 study [91]. For Western countries, the European Organisation for Research and Treatment of Cancer (EORTC) has defined the indications for endoscopic resection for EGC during the St. Gallen international consensus meeting. For diffuse EGC, gastrectomy is considered mandatory [92]. In the NCCN and ESMO guidelines undifferentiated tumors (including PC-GC) are contra-indicated for endoscopic treatment [80].

Surgery

Multiple studies have demonstrated a higher risk of positive resection margins due to the specific infiltrative characteristics of PCC-GC and a higher risk of lymph node involvement [6,8,44]. Consequently, some surgical specificities should be proposed.

According to the JCGA, a proximal margin of 5 cm is recommended in case of AGC with an infiltrative growth pattern (i.e. PCC-GC). A frozen section is advisable in case of doubt. For EGC, a gross resection margin of 2 cm should be respected [91]. A margin of 4 cm is recommended by the NCCN regardless of histological type [80]. According to the ESMO guidelines, a subtotal gastrectomy (SG) is indicated if a macroscopic proximal margin of 5 cm can be achieved. For diffuse GC and consequently for PCC-GC, a margin of 8 cm should be respected. If not, a total gastrectomy (TG) is advised [79]. In case of an antropyloric location of PCC-GC, a frozen section of the distal margin should be proposed, since there is a significant risk of duodenal invasion due to submucosal and subserosal spreading of the tumor [49].

Neither JCGA, nor ESMO or NCCN guidelines advocate a modification of the D2 Lymphadenectomy without systematic splenectomy for AGC in PCC-GC [79,80,91]. Only the guidelines of the Italian Research Group for Gastric Cancer (GIRCG) recommend a D2+ lymphadenectomy (D2 + stations 8p, 12p/b, 13, station 14 v along the mesenteric vein and para-aortic lymph node station 16a2/16b1) for tumors classified as diffuse type according to the Laurén classification and located in the distal two-thirds of the stomach [93].Whether or not the extent of lymphadenectomy should be adapted to the higher potential of lymph node metastasis in PCC-GC is questionable and has so far not been investigated by any RCT.

Impact of PCC-GC in peri-operative chemotherapy

In Western countries, before the FLOT era

The added value of perioperative CT for GC has been demonstrated in two randomized trials [17,94,95]. Perioperative CT allows for an increased R0-resection rate, tumor- and lymph node downstaging as well as significant improvement in OS. In a post hoc analysis of the MAGIC trial, no statistically significant difference in pathological response rate could be identified between the different histological types according to the Lauren classification. Of note, only 18 % of included patients presented with diffuse type GC and the presence of SRC was not specifically evaluated [96]. Other studies, mainly retrospective in nature, have suggested that Laurén diffuse type GC and SRC-GC specifically, were less chemo sensitive compared to other histological subtypes [8,97–100]. In a large multicentric retrospective cohort study among 1050 patients with SRC-GC defined as tumors with >50% SRC [13], Messager *et al* found that perioperative CT (ECF or 5FU/Cisplatin) did not result in tumor- or lymph node downstaging, nor did it entail any benefit in terms of R0 resection [100]. Perioperative administration of CT was even identified as an independent factor op poor prognosis in the SRC-GC group (HR = 1.4, 95%CI [1.1-1.9]). Several hypotheses could account for these findings: (i) innate chemoresistance of SRC-GC, (ii) disease progression during neoadjuvant CT or (iii) toxicity resulting in relative immunodepression with subsequent facilitation of disease progression [101]. The results found by Messager *et al* highlighted the urgent need of a

randomized controlled trial dedicated to the identification of an optimal therapeutic strategies in the management of SRC-GC. In this context, the phase II/III PRODIGE 19 randomized controlled trial was designed in order to evaluate whether upfront surgery with adjuvant CT (6 cycles of ECF regimen) would provide a survival benefit compared to perioperative CT (perioperative ECF regimen) in patients with stage Ib-III SRC-GC [102]. The phase II study met its primary endpoint of > 26 mo of 2-year OS in the upfront surgery + adjuvant CT arm. However, 2 year-OS rates was 60% in the perioperative arm *vs* 53.5% in the upfront surgery arm with a median survival of 39 mo vs. 28 mo respectively (exploratory hazard ratio, 0.71 [95%CI: 0.40-2.64]). Subsequently the phase III was not launched [18].

Another retrospective study, including 235 patients with SRC-GC defined as tumors with any percentage of SRC, suggested that SRC-GC had a lower clinical (21.1% vs. 33.7%, P = 0.001) and histopathological (16.3% vs. 28.9%, p<0.001) response rate to neoadjuvant CT than non SRC-GC [67]. Within the cohort of SRC-GC patients that displayed a clinical or histopathological response however, the outcome was favorable, which led to the conclusion that perioperative CT should not be abandoned for SRC-GC. In the same study, addition of a taxane-based CT regimen did not have any positive influence on prognosis in SRC-GC patients.

In Western countries in the FLOT era

Taxane-based CT regimens, and more specifically the FLOT (5-FU, Leucovorin, oxaliplatin and docetaxel) regimen, have in recent years proven their added value in the peri-operative treatment of GC [17,103,104]. Results concerning the benefit of the FLOT regimen in the treatment of PCC-GC remain however controversial: Homan *et al* found that the pathological complete response rate to FLOT therapy in intestinal type GC was higher as compared to diffuse/mixed type CG (30.8% vs. 0%, p<0.05) [105]. Likewise, in the phase II NeoFLOT study, it was demonstrated that when considering near complete responders (<10% residual tumor), 85% had an intestinal type GC in contrast to only 10% and 5% of these patients that exhibited a diffuse and mixed type tumor, respectively [106]. The results from the FLOT4 trial however demonstrated a beneficiary

treatment effect of the FLOT regimen *vs* ECF regardless of histological type and presence of an SRC component [17]. However the definition of SRC in the FLOT trial was presence of any SRC in the pathological report which does not correlate with the recent definition of PCC-GC [12]. The beneficial effect on OS was more pronounced in the SRC-GC than in diffuse GC. These findings are difficult to analyse in the absence of pathological reassessment of the pathological specimen. This was however an additional argument not to launch the phase III of PRODIGE 19 trial.

In Eastern countries

In Eastern countries where primary surgery followed by adjuvant chemotherapy is the standard treatment, thre trials evaluating preoperative chemotherapy dedicated to linitis plastica have been identified [107-110]. The first study with S1 (JCOG02) did not reach its expected survival rate and consequently no phase III study was performed, the second study with S1+ cisplatin showed interesting tumor response (JOG0210) but did not show any superiority of the neoadjuvant arm in the long term in the phase III (JCOG0501) .

Impact of PCC-GC on adjuvant chemotherapy

In Eastern countries, adjuvant CT is the preferred therapeutic strategy in GC based on 2 major trials : the ACTS-GC (Adjuvant Chemotherapy Trial of TS-1 for Gastric Cancer) trial and the CLASSIC sudy with CAPOX [111,112]. There was no subgroup analysis based on diffuse or SRC-GC type in both trials. However, in the ACTS-GC trial, S-1 setting had a significant favourable HR for death in the undifferentiated group (that includes PCC-GC) compared to surgery alone contrary to the differentiated group, where the effect was not significant [111]. After 5 years, the results were maintained in both subgroups [113]. A retrospective study suggested no tumor response of SRC-GC to either oxaliplatin or docetaxel adjuvant based chemotherapy, whereas the mixed SRC-GC group responded to both regimens with even better improved survival with the docetaxel-based regimen [98]. Although the exact definition of SRC-GC and mixed SRC-GC was not mentioned in this study, it supports the fact that PCC-GC could behave

differently according to the percentage of SRC and underlines the potential benefit of taxane-based CT in PCC-GC.

Impact of PCC-GC on adjuvant radiotherapy

Several RCT evaluated the potential benefit of adjuvant CRT in GC (Intergroup 0116, ARTIST,ARTIST2, CRITICS) [114-119]. All of them failed to show a favorable outcome in PCC or diffuse GC subgroups . An analysis of the SEER database using a propensity score however showed favourable outcome of adjuvant RT in patients with diffuse-type GC (median survival time: 30 mo with adjuvant RT vs. 18 mo without adjuvant RT, p<0.001, HR: 0.75, p<0.001). A major bias was the absence of knowledge regarding the use of CT [120].

Impact of PCC-GC on neo adjuvant chemoradiotherapy

Phase III trials evaluating RT or preoperative CRT in GC, excluding the GEJ, are few and small [121–123]. Several phase II trials showed encouraging results in terms of tumor response and survival but this type of strategy has up to now been limited by the toxicity caused [124–128]. At least two trials are in course: TOPGEAR [129] and CRITICS-II [130] with a planned subgroup analyses according to histological type only in the CRITICS-2.

A study analysing 107 Localized GA (n = 45 non-SRC-GC and n = 62 SRC-GC) treated with preoperative CRT showed presence of SRC was associated with a lower rate of pCR (11% *vs* 36%, P = 0.004) and the association remained significant even with low percentage of SRC (1–10%; P = 0.014). Higher the fraction of SRC, the lower was the probability of pCR (P = 0.03). Poorly differentiated and SRCC led to shorter OS (P = 0.046 and P = 0.038, respectively) [97].

Impact of PCC-GC in Intraperitoneal chemotherapy (IPC) combined with surgery

Preventive setting

The high failure rate of surgical curative therapy for GC and PCC-GC in particular, is mainly due to a high rate of peritoneal recurrence. In this context, a strategy of preventive IPC at the time of surgical intervention has been hypothesized. Two meta-analyses (with mostly Asian studies) showed a clear benefit of preventive IPC in terms

of survival [131,132]. However, no subgroup analysis for PCC-GC was performed. The phase III GASTRICHIP trial (NCT01882933) is currently evaluating the role of oxaliplatin-based heated intraperitoneal chemotherapy (HIPEC) in addition to curative gastrectomy in patients with GC or Siewert II/III cardia adenocarcinoma with either serosal infiltration, LN positivity, positive peritoneal cytology or perforated tumor. A stratification according to presence of SRC on pretherapeutic biopsies, has been anticipated [133]. The currently ongoing PREVENT trial (FLOT-9) (NCT04447352) is a multicenter, randomized, controlled, open-label study including a total of 200 pts. with localized and locally advanced non-metastatic diffuse or mixed type (Laurens's classification) adenocarcinoma of the stomach and Type II/III esogastric junction tumors. Patients undergo perioperative FLOT and are randomized between curative gastrectomy alone and curative gastrectomy + intra operative cisplatin based HIPEC [134]. In Japan the PHOENIX-GC2 Trial will evaluate the impact of IPC as adjuvant or perioperative chemotherapy for patients with type 4 scirrhous gastric cancer in addition to S1 chemotherapy [135].

Curative setting

In a curative setting, cytoreductive surgery (CRS) plus HIPEC has been strongly recommended for AGC by a panel of international experts [136,137]. However, controversy concerning this topic remains with further high-quality evidence being awaited to confirm the value of this treatment strategy, which could be of particular interest for PCC-GC.

At present, no published RCT has compared CRS+HIPEC vs. CT alone. Two ongoing randomized phase III trials evaluate the role of surgery in limited- metastatic adenocarcinoma of the stomach or esophagogastric junction in patients responding to chemotherapy and will include patients with peritoneal carcinomatosis [138,139]. In the RENAISSANCE trial no stratification on histological type has been anticipated and HIPEC is not described in the protocol (NCT02578368) [138].In the SURGIGAST trial, stratification on histological type (PCC-GC on biopsy) has been anticipated (NCT03042169) [139].

In the multicenter, open-label, phase III PERISCOPE II trial patients with peritoneal metastasis are currently randomized between CT alone vs. CRS+HIPEC with CT. Study completion is expected by October 2022 [140]. A stratification on the main histological subtype (diffuse vs. intestinal) has been anticipated.

Based upon the available evidence, it is presumed that for GC in general, only patients with a Peritoneal Cancer Index (PCI) < 12, who display a clinical response after neoadjuvant CT and in whom no diffuse bowel involvement is found, may benefit from the added value of CRS+HIPEC [141,142]. For PCC-GC, little to no specific selection criteria have been proposed so far. In a retrospective study on 89 patients, Chia *et al* demonstrated that after treatment with CRS+HIPEC, non PCC-GC patients had a better OS (21.8 vs. 13.2 mo, $P = 0.0214$) compared to PCC-GC patients. The authors suggested that if complete CRS was achievable in patients with a PCI<7, the presence of a SRC component should not be considered as a contra-indication for CRS +HIPEC [143].

In 2018, Bonnot *et al* published the results from the large multicenter retrospective CYTO-CHIP study, which evaluated the survival results of CRS compared to CRS + HIPEC in patients with AGC with peritoneal involvement [144]. Only patients with a complete CRS (CC-0 or CC-1) were included in the study. After propensity scored weighting, the results of this study showed that CRS+HIPEC was associated with an increased OS and potential of disease eradication as compared to CRS alone. A subgroup analysis confirmed the superiority of CRS+HIPEC in patients with PCC-GC defined according to WHO classification [11]. An ancillary study recently published showed that PCC-GC was associated with poorer OS (HR 0.43, $P = 0.003$), as were pN3, Peritoneal Cancer Index (PCI), and resection with a completeness of cytoreduction score of 1, whereas HIPEC was associated with improved OS (HR 0.52; p<0.001). The benefit of CRS-HIPEC over CRS alone was consistent, irrespective of histology, with a median OS of 16.7 *vs* 11.3 mo (HR 0.60, $P = 0.018$) in the PCC-GC group, and 34.5 *vs* 14.3 mo (HR 0.43, $P = 0.003$) in the non PCC-GC group. Non PCC-GC and HIPEC were independently associated with improved recurrence-free survival and fewer peritoneal recurrences. In patients who underwent HIPEC, PCI values < 7 and < 13 were

predictive of OS in PCC-GC and non PCC-GC populations respectively [145]. Consequently, those patients should be well-selected in order to maximally avoid the morbidity rate associated with an unnecessary exploratory laparotomy [146].

Role of PCC-GC on non curative treatments

Chemotherapy

Several studies demonstrated that SRC-GC had different infiltrative and metastatic mechanisms than non-SRC-CG. It lacked free ribosomes but were rich in lysosomes and mucus impeding anticancer drug to get to the cell [20,147]. In a metastatic setting there are few data concerning chemosensitivity of PCC-GC. Rougier *et al* reported among 87 patients with metastatic or recurrent tumor (n = 57) or with locally AGC (n = 30) a significantly poorer response rate of CT using infusional 5-FU and cisplatinum for linitis plastic or SRC histology (P = 0.003 and P = 0.16, respectively) [148].

A retrospective analysis of the FLAGS trial suggested that survival was improved among patients with advanced diffuse GC treated with S-1 and cisplatin compared to 5-FU and cisplatin [149]. A dedicated phase III trial evaluated compared both regimen in patients with metastatic diffuse gastric and GEJ adenocarcinoma previously untreated [150]. However both regimen were similar in efficacy and safety and the primary end point was not met. A study of the AGEO evaluated the place of docetaxel added to 5-FU, leucovorin and oxaliplatin (TEFOX) as first-line treatment in 65 patients with metastatic ou locally advanced non-resectable gastric ou GEJ SRC-GC including 17 LP. This regimen gave an interesting response rate of 66% with an OS of 14.3 mo. Interestingly, 26 patients (40%) initially unresectable had secondary resection (n = 24) or radiotherapy (n = 2) with curative intent [151].

Pressurized Intraperitoneal Aerosol Chemotherapy (PIPAC)

Pressurized intraperitoneal aerosol chemotherapy (PIPAC) is a recently developed promising technique which allows for homogoneous loco-regional application of intraperitoneal chemotherapy at lower doses that achievable in conventional HIPEC [152]. This technique could offer a valuable alternative for patients with unresectable peritoneal disease from gastric cancer and with PCI-scores which are considered as to

high for CRS+HIPEC (PCI>7 or 12 depending on histological type). Several retrospective studies have evaluated the feasability of this technique on patients with unresetable peritoneal metastasis from gastric cancer. The majority of patients included in these studies were affected by a SRC histology and the results show that PIPAC treatment (with low-dose cisplatin + doxorubicin) is associated with improved survival, without comprimising the quality of life [152–154]. Further results from the randomized controlled multicenter phase II PIPAC EstoK 01 trial evaluated the interest of PIPAC in addition to intravenous chemotherapy are awaited [152].

Targeted drugs in gastric SRCC

Due to some specific ongogenic pathways in GC, the efficacy of several targeted agents has been tested in recent trials, in which SRC histology has only rarely been the subject of subanalysis. Diffuse type GC on the other hand, has been evaluated frequently within these trials.

HER2 targeting agents

The incidence of human epidermal growth factor receptor 2 (HER2) amplification in GC ranges from 11% to 22.1%. It is more often noted in intestinal GC than diffuse type GC and characterized by a more frequent location in the proximal stomach and gastro-esophageal juntion [155–160]. Altough still controversial, a HER2 positive status is in general associated with poor outcome and more aggressive disease [155,157,160]. Some authors found that the unfavorable prognostic value of HER2 positivity was present in intestinal type GC, but not in diffuse type GC [161,162]. In PCC-GC, the diagnosis of HER2 status can be somewhat troublesome due to the presence of a marginalized cytoplasm and nucleus, entailing a frequent misinterpretation of strong, non-specific staining [163–165]. The phase III ToGa trial demonstrated the added value of the humanized monoclonal antibody against HER2 (Trastuzumab) in combination with CT (capecitabine or 5-FU and cisplatin) compared to CT alone in HER2-positive AGC [159]. Of note, a sub-group analysis among patients wit a diffuse-type tumor showed no benefit of trastuzumab, although the number of patients in this sub-analysis was quite low. A Korean study found a resistance to trastuzumab of more than 50% among 13

patients with SRC-GC who were HER2 positive, with a low HER2 amplification index being identified as an independent molecular predictor for trastuzumab resistance in a multivariate analysis [166]. Despite these findings, it remains nevertheless recommended to routinely test all patients with GC for HER2 amplification, regardless of the histological type [158,159,167]. Future studies are required to investigate more profoundly a potential benefit of trastuzumab in PCC-GC.

Anti-angiogenic agents

The randomized phase III AVAGAST trial evaluated the effect of bevacizumab (a humanized anti-VEGF monoclonal antibody) in combination with CT (fluoropyrimidin-cisplatin) as a first-line therapy in AGC. Although AVAGAST did not reach its primary objective (OS of 10.1 mo in the placebo arm vs. 12.1 mo in the bevacizumab arm, P = 0.1002), the addition of bevacizumab to CT was found to be associated with a significant increase in progression-free survival (PFS) and overall response rate [168]. An additional analysis according to disease subtype, suggested a benefit of bevacizumab in a subset of non-Asians patients with diffuse histologic type (HR=0.68; 95%CI[0.48-0.97]) [168]. The phase III REGARDS trial compared ramucirumab (an anti-VEGF-R2 antibody) *vs* best supportive care after first-line platinum-containing or fluoropyrimidine-containing CT in AGC or gastro-esophageal junction adenocarcinoma. Ramucirumab provided a significant benefit in terms of OS (5.2 vs. 3.8 mo, HR=0.78, 95%CI [0.603-0.998]) [169]. In subgroup analysis, a significant benefit was found for diffuse type GC (HR = 0.56; 95%CI[0.36-0.85]), but not for the intestinal type (HR = 1.009, 95%CI: 0.583-1.745), suggesting a higher sensitivity to anti-angiogenics. Conversely, the RAINBOW trial showed that for ramucirumab in combination with paclitaxel in a second line treatment, the OS benefit concerned only the intestinal histological subtype (HR: 0.705 (0·534–0·932) [170]. Supplemental data are needed to establish the role of anti-angiogenic targeted therapies in patients with diffuse type GC. At present, no data concerning the role of anti-angiogenic therapies in the therapy of PCC-GC are available.

Anti- EGFR (epidermal growth factor receptor)

EGFR expression has been identified as an independent predictor of poor prognosis in patients with PCC-GC compared to non PCC-GC patients [171]. Data from the EXPAND and REAL3 trials have suggested no additional benefit of anti-EGFR treatment in combination with CT for AGC [172,173]. In a subgroup analysis of the EXPAND trial in function of the histological subtype, it was even found that anti-EGFR could be harmful in diffuse type tumors (HR for OS: 1.44, 95%CI [1.01-2.03]) [172].

Mammalian target of rapamycin (mTOR) inhibitors

Since phospho-mTOR is expressed in 60% of intestinal and 64% of diffuse-type GC, mTOR inhibitors were considered as an interesting therapeutic option from a biological point of view [174]. Results from the phase III GRANITE-1 trial however showed no benefit of everolimus (an oral mTOR-inhibitor) on OS compared to best supportive care for previously treated AGC [175]. In a subgroup analaysis, no benefit in diffuse-type GC was found either.

CLDN18.2 antibody (Zoltemuximab)

In advanced gastric/gastro-oesophageal junction and oesophageal adenocarcinoma patients expressing CLDN18.2, adding zolbetuximab to first-line EOX provided longer PFS and OS *vs* EOX alone in a phase 2 trial [176]. Interestingly the vast majority of these population had diffuse of mixed type GC. Zolbetuximab is being evaluated in phase III studies based on clinical benefit observed in the overall population and in patients with moderate-to-strong CLDN18.2 expression in >70% of tumour cells.

Immunotherapy

Among new treatment strategies for GC, immunotherapy, and more specific PD-L1 (programmed death-ligand1) inhibitors have proven to be the most promising. PD-L1 is expressed in 30% to 63% of GC [177,178]. The results of the CheckMate 649 study, demonstrated the superiority of nivolumab in combination with CT compared to chemotherapy alone. In a study population of patients with HER2 negative, previously untreated, unresectable advanced, or metastatic GC or gastro-oesophageal junction cancer, nivolumab in combination with CT (XELOX or FOLFOX) resulted in significantly improved OS and PFS *vs* chemotherapy in patients whose tumors

expressed a PD-L1 combined positive score (CPS) ≥ 5 (HR for OS= 0.71, 98.4% CI [0.59–0.86] and HR for PFS= 0.68, 98% [CI 0.56–0.81]). This survival benefit was also observed in the group of patients with a PD-L1 CPS ≥ 1 and in the all-randomized population [179]. The rate of patients with SRC-GC or diffuse tumors was close between patients with a CPS≥ 5 and the overall population [179]. However other studies found that in SRC histology, PD-L1 CPS >1 was significantly less observed [180]. The question remains how the recent findings of the CheckMate 649 trial could be applied to PCC-GC. A group of specifically selected PCC-GC patients with SI may benefit from immunotherapy. However, Hirotsu *et al* reported that PCC-GC exhibits high MSI at low frequencies [181].

CONCLUSION

In contrast to GC in general, the relative incidence of PCC-GC has risen over the past few decades. PCC-GC represents a distinct pathological entity within the GC spectrum, characterized by specific epidemiological and clinical features, including younger age at presentation and a significantly worse prognosis, mostly due to peritoneal dissemination early on in the disease. In light of these distinct features, the recently redefined pathological definition of PCC-GC by the WHO and the European chapter of IGCA will facilitate methodological standardization in future studies, which in turn will help to identify which therapeutic strategies for GC in general are applicable to PCC-GC. We believe that the updated definition will help standardize future research concerning the prognostic results of SRC-ECG in Western populations as well as in evaluating the correlation between pre-therapeutic biopsis and the final pathology result. Concerning the pre-therapeutic evaluation, the infiltrative growth pattern of PCC-GC along with early peritoneal dissemination justifies the use of repeat endoscopies with deep biopsies, CT-graphic imaging as well as systematic staging laparoscopy with peritoneal lavage. Since correct PCI determination is essential for therapeutic management, a small incision with palpation of the entire small bowel shoud be considerd. Surgery is considered the mainstay of curative treatment for PCC-AGC. The role of the extent of the lymphadenectomy however in PCC-AGC should be

evaluated in future studies. For PCC-EGC, no endoscopic treatment is currently advocated. The added value of peri-operative CT for PCC-GC with FLOT regimen is probable but should be further confirmed using histological reassessment. No role of adjuvant radiotherapy has been demonstrated in PCC-GC. In case of peritoneal disease, IPC by means of HIPEC, PIPAC offer a valuable treatment option, on the condition that patients are well selected. To what extent the promising results of immunotherapy could be applicable to PCC-GC needs to be confirmed in future studies. PCC-GC in general requires a highly individualized diagnostic and therapeutic approach to optimize the inherent poor prognosis of this disease in the future. Molecular and genetic differentiation will be of importance to offer a patient tailored therapeutic strategy.

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