Evaluation of different methods to assess homologous recombination status and sensitivity to PARP inhibitors in ovarian cancer

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Methods: Matched samples of ascites and tumor tissue were taken from patients undergoing surgery for epithelial ovarian cancer. Tumor samples were formalin fixed and paraffin embedded (FFPE); ascites samples were used to generate primary cultures (PC). HR status was determined in PCs as previously described. IC_{50} for the PARP inhibitor Rucaparib was estimated using SRB assays. DNA was extracted from the FFPE tissue. The following techniques were evaluated in PCs or paired FFPE samples: DR-GFP reporter assay, PARP activity assay, BRCA1 expression on immunohistochemistry, BRCA1 methylation status and BRCA1/2 mutation analysis. A next generation sequencing based assay was used to detect mutations and other genomic alterations in a large panel of cancer-associated genes, including BRCA1/2.

Results: Paired samples were collected from 64 patients and characterized for HR function. 47/64 (76%) were high grade serous. 44% (28/64) were HR defective (HRD) by Rad51 assay and correlated with Rucaparib sensitivity (PPV-92%, NPV-100%). Molecular analysis revealed that all mutations and other genomic alterations detected in ascites derived PCs were also found in matched FFPE tumor tissues. All tumors with serous histology contained p53 mutations, whilst the remaining tumors without p53 mutations were non-serous in histology. DR-GFP assay was unreliable in PC due to poor transfection. In a subset of 50 cancers there was reduced BRCA1 expression in the HRD vs. HRC tumours (34.8% vs. 22.7%, ns) whilst in a further subset of 30 cases there was no difference in endogenous or stimulated PARP activity between HRD and HRC tumours. Deleterious BRCA2 mutations were identified in 7 tumors, 6 of which were HRD. Only 1 deleterious BRCA1 mutation was detected but methylation of BRCA1 was identified in 13 of 64 (20%) tumors, 7 of which were HRD. Mutation of BRCA2 was mutually exclusive to methylation of BRCA1. HRD vs. HRC tumours showed BRCA1 methylation (25% vs. 17%) and BRCA1/2 mutation (21% vs. 0.3%). 14/28 (50%) HR defective tumors do not have BRCA1/2 mutations or BRCA1 methylation, suggesting other mechanisms can also result in a HR defective phenotype. 28/64 (43%) of samples demonstrated the HR defective phenotype. In all cases HR status was correlated with sensitivity to Rucaparib. Conclusion: As expected, deleterious BRCA2 mutations conferred a HRD phenotype in cells but methylation of BRCA1 was not universally associated with HRD. This may be as a result of only partial silencing of the gene by methylation and further work is required to identify thresholds of methylation which may predict HR status. The use of BRCA1/2 mutation testing alone is unlikely to identify the majority of HR defective ovarian tumors. Assessment of functional status of HRD is the preferred option and further technologies should be developed to simplify the Rad51 assay.

Evaluation of supragastric lesser sac using a laparoscope during cytoreductive surgery in epithelial ovarian carcinoma: A site for occult metastasis

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Background: The supragastric lesser sac (SGLS) is a site of metastasis from epithelial ovarian cancer (EOC). Since this region is difficult to access and represents a confluence of critical structures, it may be a barrier to complete cytoreductive surgery (CRS).

Methods: The SGLS was explored in consecutive patients undergoing CRS with EOC. After a xipho-pubic laparotomy incision, the SGLS was examined; visualization and treatment was aided by using a laparoscope. Resectable disease was cleared using the following methods alone or in combination: direct tumor excision, argon beam coagulation, plasma jet or electrocautery.

Results: 30 patients were evaluated between November 2013 and August 2014 in NGOC, Gateshead. SGLSM was present in 21/30 (70%) of EOCs, 19/25 (76%) high grade serous disease, 21/26 (81%) stage ≥3 disease, 18/20 (90%) with PCI score ≥15, 12/15 (80%) with ascites ≥500 ml, 13/18 (72%) at primary surgery and 8/10 (80%) at interval surgery. Sites included: lesser omentum (11), caudate lobe (10), groove of ligamentum venosum (6), floor (20), upper recess (7), subpyloric space (6), FOW (13), coeliac axis (5), portal hepatitis (6), anterior surface of pancreas (2) retro-pancreatic (2). Size of metastases: <2.5mm = 3, <1 cm = 8, ≥1 cm = 7. Pre-operative CT scan identified 4/22 (18%) cases. In 18/21 patients SGLSM was completely resected or ablated; there were no complications. End Result: Optimal 27/30 (90%) including no visible disease = 18, <2.5 mm = 5, 17/21 (81%) cases would have been ≥2.5 mm residual disease if SGLSM was not evaluated/treated. In a further cohort of 30 patients evaluated at Tata Medical Center, Kolkata, SGLSM was present in 18 (60%) of patients. CC1 resection was obtained in >90% cases.

Conclusion: EOC frequently metastasizes to the SGLS and is often resectable. Lack of meticulous examination may result in incomplete resection; evaluation should be performed at least in stage ≥3 disease when the surgical intent is total clearance of disease.

Implementing quality indicators for cytoreductive surgery in ovarian cancer: Experience from a tertiary referral center in Eastern India

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Background: Debate continues whether primary surgery or neo-adjuvant chemotherapy (NACT) or primary debulking surgery (PDS) should be offered in advanced epithelial ovarian cancer as frontline therapy. Since 2015, there has been a paradigm shift at Tata Medical center, whereas increasing number of patients are being offered PDS and a quality improvement programme is initiated. Recently, ESGO in October 2015 has published a document indicating 10 quality indicators for cytoreductive surgery in advanced ovarian cancer surgery.

Aim: We compared our performance against all 10 quality indicators.

Results: Primary cytoreduction rate has increased from 20% in 2012 to >70% at the end of 2015. Optimal cytoreduction rates were obtained in 90% cases and recently complete (CCO/CC1) cytoreduction rates are being achieved in >80% cases. All 10 quality indicators were achieved successfully including prospective documentation of morbidity and surgical findings in all cases. Morbidity figures are showing a downwards trend after the initial learning curve.

Conclusions: Implementation of a quality improvement programme is the key to overcome the barriers of implementing a cytoreductive program in advanced ovarian cancer. However, standards similar to developed countries can be achieved through a dedicated team effort.
Background: TCGA data using expensive multi-modality diagnostic platforms have shown that 50% of epithelial ovarian cancers (EOCs) are estimated to be homologous recombination (HR) deficient (HRD). We developed a functional assay for HR using gamma H2AX-Rad51 immuno-fluorescence.[1]

Methods: Primary cultures were developed in 50 consecutive EOCs from ascitic fluid and HR assay was performed.

Results: 50% patients were HRD based on the functional assay and show improved ex-vivo chemosensitivity to PARP inhibitor (PARPi) (PARP1) (PPV = 92%, NPV = 100%). HRD patients showed improved platinum sensitivity (53.8% vs 16.7%), survival (12 month OS - 41.7% vs. 11.5%) and optimal cytoreduction (80% vs. 62%) rates compared to HR competent (HRC) tumours which are less responsive and represent an unmet clinical need.

Conclusions: Personalised surgical and chemotherapeutic strategies may be developed for HR stratified EOCs. Primary surgery may be the preferred approach in HRC due to poor chemoresponse; surgical expertise/ environment should be optimised to ensure optimal surgical outcome. Intra-operative hyperthermic treatment and selective HR inhibitors may improve subsequent chemoresponse in HRC and are currently being investigated.

Reference:

Ovary: Oral Abstract

Diagnostic accuracy of intraoperative frozen section in ovarian neoplasms: Experience in a tertiary oncology centre

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Aims and Objectives: This study is done to assess the accuracy of intraoperative frozen section in the diagnosis of various categories of ovarian neoplasm conducted in RGCI.

Materials and Methods: Intraoperative frozen sections for suspected ovarian neoplasm that underwent surgery as primary line of therapy at this institution were analyzed retrospectively from Jan. 2014 - Dec. 2015. The results of frozen section were compared with the final histopathologic diagnosis on paraffin sections and the overall accuracy, sensitivity, specificity, positive and negative predictive values were determined.

Results: The study included 159 cases and the mean age of patients was 44.72 ± 14.28 years (Range 19-75 years). The mean size of tumor was 12.5 ± 5.9 cm. Sensitivity of frozen section for benign, borderline and malignant tumors was 98.53%, 73.33% and 94.74% respectively. And the related specificities were 95.68%, 96.53% and 100% respectively. There were 150 concordant cases and 9 discordant cases. Overall diagnostic accuracy of frozen section was 94.33%.

Conclusion: Intraoperative frozen section diagnosis appears to be an accurate and comparable technique for the histopathologic diagnosis of ovarian tumours. It is a valuable tool to guide the surgical management of these patients.

Key words: Frozen section; ovarian neoplasm

Ovary: Oral Abstract

Modified posterior pelvic exenteration and rectosigmoid anastomosis for advanced epithelial ovarian cancer: A safe cytoreductive procedure

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Introduction: Surgery plays an important role in the management of advanced stage ovarian cancer and is complex involving surgical procedures including peritoneectomy, splenectomy, diaphragmatic stripping, retroperitoneal lymph node dissection and bowel resection including resection of recto-sigmoid.

Objective: To assess the safety and efficacy of the patients undergoing modified posterior pelvic exenteration and rectosigmoid anastomosis achieving in optimal cytoreduction.

Methods: Between June 2011 and June 2014 a total of 100 patients underwent surgical cytoreduction for advanced epithelial ovarian cancer of which 20 patients had undergone modified posterior pelvic exenteration with rectosigmoid anastomosis. The present study includes a retrospective analysis of these 20 patients. Rectosigmoid anastomosis was done using circular stapler in these patients. All patients had a PS score of 1 or 2.

Results: The median age of patients was 50 years. The optimal status of no macroscopic residual disease was achieved in all patients. Modified posterior pelvic exenteration with rectosigmoid anastomosis was carried out to achieve optimal status of surgical cytoreduction in 20 patients out of which fifteen patients had primary surgical cytoreduction, three patients had interval surgical cytoreduction surgery after receiving three cycles of neoadjuvant chemotherapy with paclitaxel & carboplatin while two patients had this procedure as a part of secondary surgical cytoreduction. The most common histology was papillary serous carcinoma. Average blood loss was 500 ml. Mean operative time was 6 hours. There were no intra operative complications. Bowel movements returned to normal in 3 to 5 days. The median length of hospital stay was 7 days. The median time to start post-operative chemotherapy was 32 days. There was no major morbidity and mortality.

Conclusion: Modified posterior pelvic exenteration with rectosigmoid anastomosis should be performed when indicated as a part of cytoreduction. In our experience this is a safe and effective procedure to achieve optimal status in advanced ovarian cancer.

Ovary: Oral Abstract

Evaluation of ovarian reserve in women undergoing ovarian cystectomy by laparoscopy and laparotomy

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Ovarian cysts are one of the commonest problems encountered in the gynecological field. Majority of these cysts are functional i.e., disappear spontaneously, while few need cystectomy. Ovarian cystectomy is done by laparotomy and laparoscopic technique. The method to achieve haemostasis in the ovarian bed after cyst removal varies with the type of technique. Electrocoagulation is used to achieve haemostasis in laparoscopic cystectomy while the bleeding vessels are sutured for haemostasis in cystectomy by laparotomy. Both the modalities of management varies in terms of compromise of ovarian reserve. The study was carried out to evaluate the surgical impact of benign ovarian masses on ovarian reserve as measured by serum levels of antimullerian hormone. In this prospective study on 30 women of reproductive age group with benign ovarian masses, 15 women were enrolled for laparoscopic ovarian cystectomy and another 15 women were enrolled for cystectomy by laparotomy and ovarian reserve was measured by levels of serum AMH preoperatively, postoperative one week and postoperative 3 months using standard ELISA assay kit. The preoperative, postoperative one week and postoperative 3 months levels of mean AMH were 4.74 ± 1.86 ng/ml, 2.92 ± 1.45 ng/ml and 2.64 ± 0.96 ng/ml respectively, in women undergoing laparoscopic cystectomy and 3.98 ± 1.35 ng/ml, 2.48 ± 0.64 ng/ml and 2.11 ± 0.63 ng/ml respectively in women undergoing ovarian cystectomy by laparotomy. So there was decline of mean AMH levels in postoperative one week and postoperative 3 months samples in both of the groups of enrolled women. However, this decline varied with the type of cyst removed and it is insignificantly greater in laparoscopy group, wherein electrocoagulation may cause extensive and sustained damage to ovarian tissue.

Ovary: Oral Abstract

Multiple recurrence of granulosa cell tumor of the ovary: a case report and literature review

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