Primary Squamous Cell Cancer of the Rectum – An Update
Response to Reviewers

12th October 2015

Dear Editor,

Please find enclosed the edited manuscript in word format (file name: 21140-review.docx)

Title: Primary Squamous Cell Cancer of the Rectum

Authors: Glen R Guerra, Cherng H Kong, Satish K Warrier, Andrew C Lynch, Alexander G Heriot, Samuel YK Ngan

Name of journal: *World Journal of Gastrointestinal Surgery*

ESPS Manuscript No: 21140

The manuscript has been revised in light of the reviewers' comments.

REVIEWER **00227406**

'The review article by Guerra et al is a comprehensive overview of primary rectal squamous cell carcinoma. The layout is clear and the presentation systematic. Current controversies and emerging therapies are discussed in a balanced manner. The language is stylistically fluent and straight forward to read. It is well illustrated. However, I would ask that the images are labelled to allow non pathologists / radiologists to better appreciate the abnormalities described in the figure legends. This is my only suggestion for improvement for this paper, which provides a valuable addition to the body of work addressing this often neglected tumour type.'

RESPONSE: ***Thankyou for your positive appraisal.***
The figures have been labelled to clarify the abnormalities depicted.

REVIEWER ***00227423***

'This is an excellent review for a "rare condition". The manuscript summarized all aspects of the disease concisely and I believe this will be of great information for clinicians. If possible more explanation in precise endoscopic findings (i.e existence of ICLP) and pictures would be of help to educate clinicians.'

RESPONSE: ***Thankyou for you encouraging comments.***

There has been a single report identifying an endoscopic appearance of IPCL (intraepithelial papillary capillary loops) using NBI in an area of squamous metaplasia of the rectum (Fu, 2008). In the oesophagus, there are classification systems utilising the appearance of IPCL in order to identify and differentiate squamous lesions along the spectrum towards invasive carcinoma. This may be translatable as a means of surveillance of the rectum in high risk patients, particularly in ulcerative colitis.

Due to our limited cohort of rectal SCC patients, we have no further images available to display the endoscopic features of rectal SCC.

Endoscopic findings

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The definitive diagnosis of rectal SCC is confirmed by performing a complete colonoscopy with biopsies of any abnormalities. Demonstration of the discontinuity of a lesion from the anal squamous mucosa is of great importance. Rectal SCC has been reported to have a varied endoscopic appearance dependent on the stage of disease. This can range from a small mucosal polyp (Image 3), plaque or ulceration through to a large obstructing mass (Image 4)[1]. Pre-malignant lesions in the form of squamous metaplasia have also been identified by way of narrow band imaging (NBI) in addition to rectal SCC[2, 3]. ***One report identified an appearance of dark brown dots similar to the intraepithelial papillary capillary loops (IPCL) which herald squamous epithelium in the oesophagus using NBI (Fu, 2008)[2]. There are classification systems utilising the appearance of IPCL in the oesophagus in order to identify and differentiate squamous lesions along the spectrum towards invasive carcinoma (Boeriu, 2015)[4].*** Given the possible aetiological sequence of metaplasia through to invasive carcinoma, NBI may find a role in the detection and treatment of pre-malignant lesions for those at high risk, in particular ulcerative colitis patients.

REVIEWER ***00074724***

'In the introduction to this paper, the authors have described this work as " Systematic Review" by stating: "Furthermore, there is significant heterogeneity in the treatment regimens utilised, with the optimal management yet to be clarified. Nonetheless, certain patterns do emerge on reviewing all published cases by way of a systematic review, to determine where our future research should be directed in order to improve upon treatment and facilitate best patient outcomes." However, it is unclear how they identified the examined articles; what were the inclusion and exclusion criteria?'

RESPONSE: ***Thankyou for your useful comments.***
A methods and results section has been added to the manuscript in order to clarify the process undertaken for article selection in the qualitative and quantitative analysis.

Methods

A systematic literature review was conducted according to the guidelines of the Preferred Reporting Items for Systematic Reviews and Meta-Analyses group. A comprehensive search of Ovid Medline was performed with the abstracts screened to determine relevant articles, following which the full texts were obtained. A directed manual review of all embedded references was undertaken of the selected articles to ensure all studies published on primary squamous cell carcinoma of the rectum were identified.

The search strategy was based on a combination of medical subject heading (MeSH) terms (carcinoma, squamous cell; rectum) and text words (squamous cell carcinoma, SCC and rectum), spanning from 1946 to May 2015. The search was limited to English language with the most recent search performed on 8th May 2015.

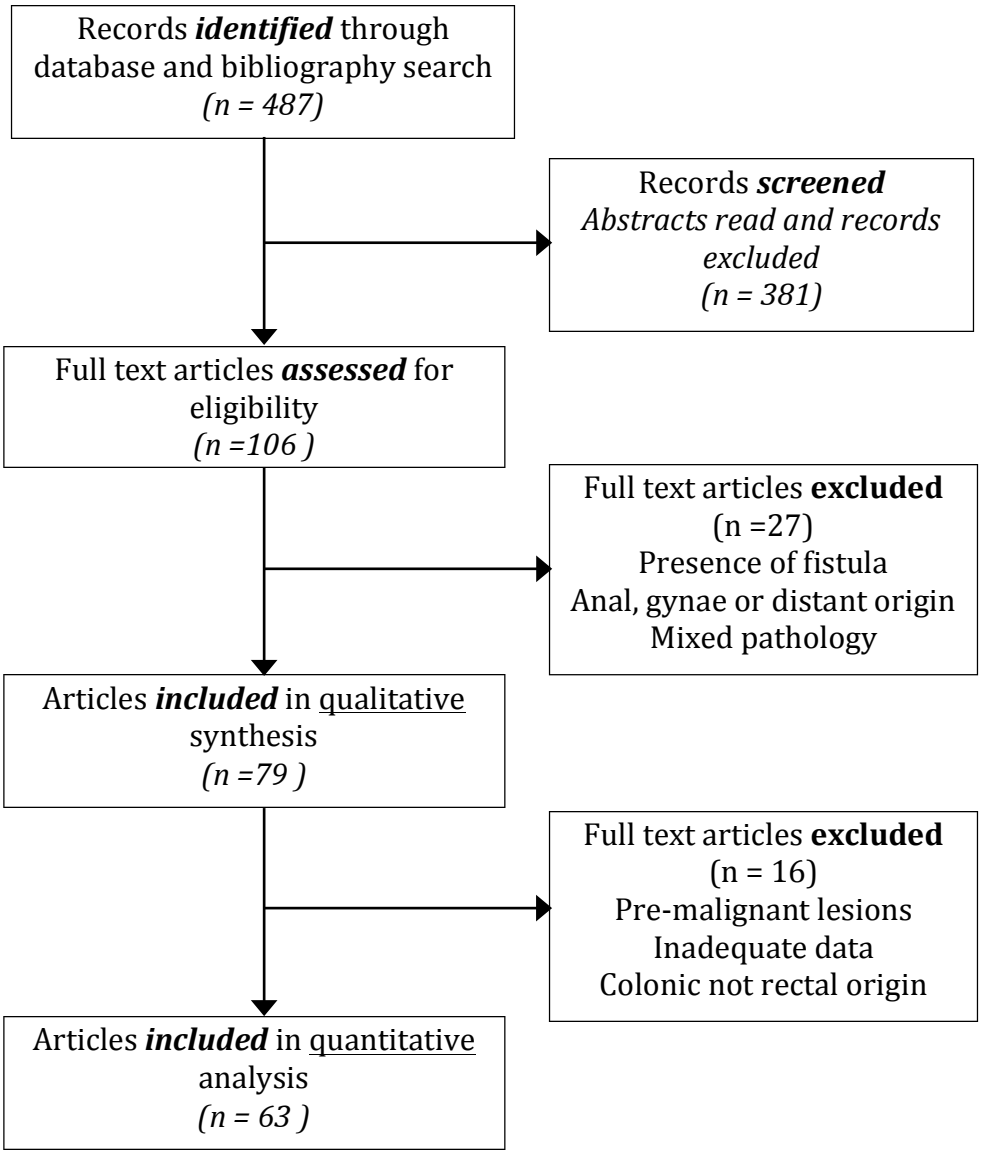

Figure 1 – **PRISMA flow diagram**.

Inclusion and exclusion criteria

The definition of rectal SCC including the three criteria as stipulated by Williams et al (detailed in 'diagnosis' below) were used to identify relevant studies. Consequently, studies reporting rectal SCC arising in the presence of a fistula, from an anal or gynaecological origin, a distant site via metastasis, or where the pathology was mixed (eg adenosquamous) were excluded. Additionally, studies where the lesion was premalignant (eg metaplasia or SCC in situ), of colonic rather than rectal origin or where the data was inadequate were exluded from the quantitative analysis.

Data Extraction

Data extracted included the names of the authors, date of publication, demographic information and clinical presentation. Location of the lesion and treatment detailing the primary modality, the use of pre- and/or post-operative modalities and the type of operation where present was also noted. Other collated information included patient outcomes in the form of local recurrence, metastasis, and survival, as well as the length of follow up. Radiological, clinical and pathological complete response was also recorded where available.

Results

The database and bibliography search identified 487 articles in total. 103 were included in the qualitative review and 63 in the quantitative analysis as detailed in figure 1. This included 43 case reports and 20 case series with a total of 142 individual cases reported.

REVIEWER ***00058510***

'An excellent review, covering this rare entity. In the section on immunohistochemistry the authors have not mentioned the uses of CK5/6 or p63, both of which have gained some popularity in this regard. In the section on treatment, the type of radiotherapy is not discussed in detail. Some have proposed contact radiotherapy (Papillon technique) and a note about whether the authors advocate this, or more standard approach with modern external beam techniques would be helpful.'

RESPONSE: ***Thankyou for your positive comments.***
CK 5/6 and p63 are useful cytokeratin markers in order to confirm carinoma of squamous origin. In the text I had listed an antibody to CK 5 (34BE12), however, I have altered this to now read appropriately.

Clinical Presentation and Diagnosis
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Histologically, if the diagnosis remains unclear, immunohistochemistry can aid in the characterisation of the lesion. This is particularly useful in cases of poorly differentiated tumours where the morphology and architecture provide little clue to the origin. Cytokeratins AE1/AE3, ***CK 5/6 (34BE12 stains CK5) and p63*** stain for cells of squamous origin, assisting in the differentiation from a rectal adenocarcinoma (Kaufmann, 2001)[5]. Cytokeratin CAM5.2 aids in the differentiation of rectal from anal, characteristically staining for rectal squamous cell or adenocarcinoma but not anal SCC. This is particularly useful for squamous carcinomas of the lower rectum[6]

There has been no reported use of contact radiotherapy (Papillon technique or contact x-ray brachytherapy (CXB)) in the literature for rectal SCC. Contact radiotherapy as a treatment in early rectal cancer has a limited evidence base. It is currently only offered to patients who are unfit for surgery with early tumours (T1N0) within close proximity of the anal canal. Given that most rectal SCC's present at a late stage and often higher in the mid or upper rectum, contact radiotherapy is unlikely to find a clear role in the treatment of rectal SCC.

Chemoradiotherapy

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Despite the encouraging results of chemoradiotherapy, currently a set treatment protocol is yet to be established. It appears that 5FU based chemotherapy combined with high dose ***external beam*** radiotherapy may be efficacious. However, while these trends are grossly evident from the literature, there is a need for further research in order to determine the most effective regimen to optimise patient outcomes.

It is unlikely that a randomised trial comparing surgery and chemoradiotherapy will ever be conducted for this rare cancer. Given the current knowledge base, it may be reasonable to suggest that primary treatment should be chemoradiotherapy, with surgery reserved as a salvage option. The suggested regimen would be a total dose of 50.4 to 54Gy ***external beam radiation*** in 1.8Gy per fraction, given concurrently with 5FU and mitomycin C.

REVIEWER ***00041966***

'This is an interesting review on squamous cells carcinoma of the rectum. The paper is well written and provides a comprehensive overview of all major issue including the therapeutic options. Among these the role of protective or definitive stoma should be mentioned. The possible association with HIV infection should also be discussed.'

RESPONSE: Thankyou for your positive feedback.

The role of a protective stoma is significant in rectal SCC in the same manner that it remains important in the management of rectal adenocarcinoma, in the setting of a low anastomosis and in patients presenting with obstruction.

While in certain cases such as low or locally advanced tumours a permanent stoma is unavoidable, the literature suggests a move towards sphincter preservation wherever possible. No information regarding continence and quality of life post treatment with sphincter preservation was offered in the literature reviewed.

Surgery

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For most Rectal SCC's, anterior resection (AR) or abdominoperineal resection (APR) has classically been performed. The choice and extent of the operation is dependent upon the tumour location and depth of invasion, occasionally requiring exenteration, with removal of involved pelvic structures. On review of the literature, APR was performed much more frequently than AR prior to the year 2000, with an equal split in the frequency of both procedures following the turn of the century (Table 1). This is likely to reflect both the change towards sphincter preservation **and avoidance of a permanent stoma** in operations for rectal cancer over previous decades, in addition to a down-staging effect of chemoradiation, which is now commonplace. ***While the incidence of APR and a definitive stoma has been falling, in a similar manner to rectal adenocarcinoma, most patients with a low rectal SCC will require a temporary covering ileostomy given the greater risk of anastomotic leak. Furthermore, for those patients presenting with an obstructing tumour, the use of a defunctioning stoma is an attractive option, providing time to appropriately stage the patient and consider the most appropriate treatment, including definitive chemoradiotherapy.***

HIV infection increases susceptibility to several virally driven cancers including the anogenital group. Consequently it seems logical that it would predispose patients to rectal SCC, however, review of the literature reveals only two reported cases. The rarity of cases in this population may be explained by the fact that most HIV patients don't develop cancers at an earlier age than the general population but have a significantly shorter lifespan, in the context of the average age of rectal SCC diagnosis being 63.

Pathogenesis

The case presented by *Sotlar et al*[7] is also of particular interest, given that it reported the findings of adjacent squamous metaplasia, dysplasia, and carcinoma in sequence, with HPV 16 identified in all three components and the surrounding non-tumour affected rectal mucosa. This mirrors the pre-neoplastic to neoplastic progression well documented in HPV driven ano-genital cancers. Furthermore, they identified transcriptional activity of the HPV E6/7 oncogenes critical to HPV's role in carcinogenesis. This may suggest that there are two possible pathways to the pathogenesis of colorectal SCC, HPV driven and non-HPV driven. However, while there is currently limited evidence surrounding HPV in rectal SCC, a clear association and a role in causation remains to be proven.

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HIV infection increases susceptibility to virally promoted cancers including Burkitt's lymphoma (Epstein Barr Virus), Kaposi's sarcoma (Human Herpes Virus 8) and anogenital carcinoma (HPV). Consequently, it could be inferred that the cell mediated immune deficiency associated with HIV would predispose to rectal SCC. However, this is not borne out on review of the literature, with only two reported cases of rectal SCC in the setting of HIV infection (Matsuda 2009, Choi 2014)[8, 9].

REVIEWER ***03305236***

'The review article described the Background, Diagnosis and Staging, Treatment, and Prognosis of Primary Squamous Cell Carcinoma of the Rectum. The whole article is well written and characterized. The article suggested to be accepted with minor language polishing required (suggested to be little simplified) . The minor suggestion: In treatment section, the authors may be better to mention whether there have any clinical study data regarding target therapy for Primary Squamous Cell Carcinoma of the Rectum.'

RESPONSE: ***Thankyou for your encouraging critique***

The manuscript language has been simplified where possible.

While there has been significant research undertaken with consequent clinical gains in the fields of molecular targeted therapy and immunotherapies over recent years, there is currently no literature in the setting of rectal SCC. Nonetheless, given the encouraging results in certain tumours, both modalities hold promise as a treatment option in the future.

Treatment

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It is unlikely that a randomised trial comparing surgery and chemoradiotherapy will ever be conducted for this rare cancer. Given the current knowledge base, it may be reasonable to suggest that primary treatment should be chemoradiotherapy, with surgery reserved as a salvage option. The suggested regimen would be a total dose of 50.4 to 54Gy in 1.8Gy per fraction, given concurrently with 5FU and mitomycin C.

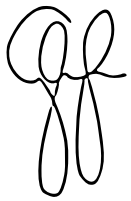
Future Options

Over recent time, there has been an increasing use of molecular targeted therapies in solid and haematological malignancies. Furthermore,

immunotherapy in the form of tumour vaccines and immune checkpoint inhibitors has become a major focus for research in the treatment of cancer with translated clinical success in specific tumour types. While there is currently no literature on these modalities in rectal SCC, the early results in other tumours holds promise for a possible role in future treatment, particularly in the cohort of patients with persistent, recurrent or metastatic rectal SCC.

Thank you,

Yours Sincerely,



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