Ocular Surface Squamous Neoplasia

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Abstract

Ocular surface squamous neoplasia (OSSN) are important because they mimic many common indolent lesions like pterygium and have a potential for causing ocular and systemic morbidity and mortality. Ultraviolet-B light and human papilloma viruses have been proposed as major risk factors in the etiopathogensis. Anterior segment optical coherence tomography, ultrasonic biomicroscopy and impression cytology have been added recently as non-surgical tools for diagnosis and management. The treatment includes surgery as mainstay with Mitomycin-C and cryotherapy as adjuvant therapy. Recently Interferons (IFN) as well as pegylated IFNs are regularly being used in a select subsets of OSSN for treatment.

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Corneal and conjunctival squamous lesions are uncommon but important because of their potential for causing ocular and even systemic morbidity and mortality. The clinical presentation of these lesions varies across a wide spectrum and can range from mild to severe dysplasia to full-thickness epithelial dysplasia (carcinoma in situ) and invasive squamous cell carcinoma. Squamous lesions can involve the conjunctiva or the cornea, but more commonly start in the conjunctiva and extend across the limbus to the adjacent cornea. Even though the first case was described in 1860 by von Graefe[1] and has been extensively described after that, the management of these lesions has changed significantly in the past decade.

The initial cases of squamous neoplasms described in the literature were cases of squamous cell carcinoma.[2] Subsequently, it has been recognized that both invasive and non-invasive subtypes of squamous neoplasms occur.[3-8] Following recognition of non-invasive forms of squamous neoplasms, various terms have been used to describe these, including epithelial plaque, Bowenoid epithelioma, and precancerous epithelioma. Pizzarello and Jakobiec[9] classified conjunctival intraepithelial neoplasms as mild, moderate and severe dysplasia based on the extent of involvement. Lesions that involved the basal one-third of the conjunctiva were classified as mild, those involving the inner two-thirds were classified as moderate, and lesions that were full thickness were termed severe dysplasia. Waring et al[10] extended the term to include the cornea, and Erie et al[11] further extended it to include invasive neoplasia.

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Ocular Surface Squamous Neoplasia (OSSN) was a term given by Lee and Hirst[12] which has three grades :-

I. Benign dysplasia

- Papilloma
- Pseudotheliomatous hyperplasia
- Benign hereditary intraepithelial dyskeratosis

II. Preinvasive OSSN

- Conjunctival/corneal carcinoma in situ

III. Invasive OSSN

- Squamous carcinoma
- Mucoepidermoid carcinoma

Incidence

OSSN is uncommon and it primarily occurs in older males (78.5%). Various authors have placed the incidence between 0.13 to 1.9/100000. It is predominantly seen in dark skinned Caucasians, the age of onset being significantly higher in areas closer to the equator. The average age of occurrence has been noted to be 60 years, ranging from 20 to 88 years.[13] The average age of incidence of carcinoma in situ lesions is 5-9 years lower than invasive OSSN. This difference represents the time taken for progression from intraepithelial neoplasm to invasive carcinoma. Patients of xeroderma pigmentosa and human immunodeficiency virus (HIV) develop OSSN at a younger age. Young patients of HIV are more prone to develop aggressive OSSN. Templeton[14] found an average incidence of 0.13/100,000 in tribal groups in Uganda. A study conducted in Brisbane, Australia found the incidence to be 1.9/100,000 population. The incidence of squamous cell carcinoma of the skin was also high, 600/100,000 in the latter geographic area. Sun and

co-workers[15] noted that the incidence was 0.3 per million per year in a study from the United States. The number of cases of OSSN relative to the total number of oculo-orbital tumours ranges from 4% to 29%. In the older population, OSSN is the third most common ocular tumour after melanoma and lymphoma.[12] Benign lesions are at least a third as frequent as malignant lesions of the ocular surface.

Etiopathogenesis

1. **Ultraviolet-B light** - UV-B light causes DNA damage and formation of pyrimidine dimers . Major risk factors for OSSN like skin colour, pale iris, sunburn, sun exposure can be attributed to UV-B irradiation. UV-B has also been shown to cause p53 gene mutation, which is associated with OSSN.[16] Histological evidence of solar injury, which is recognised as a major risk factor for conjunctival OSSN, has been reported in 50-100% cases of OSSN. Failure or delay in repair of DNA as in xeroderma pigmentosa can also lead to OSSN.
2. **Human Papilloma Virus** - HPV genotypes 6 and 11 have been demonstrated in a large number of papillomas as well as dysplastic and malignant lesions of the cornea and conjunctiva. Scott et al demonstrated HPV 16 and 18 Deoxyribonucleic Acid (DNA) and messenger Ribonucleic acid (m-RNA) in conjunctival intraepithelial neoplasia (CIN) cases proving a causal relationship. It has been demonstrated that the protein coded by the E6 region of HPV 16 and 18 forms a complex with the protein coded by the p53 tumor suppressor gene in the host.[17]
3. **Stem cell theory** - OSSN may represent the abnormal maturation of corneal and conjunctival epithelium as a result of a combination of damaging factors to the limbal transition zone such as UV-B irradiation and HPV. Other less significant risk factors include chemical exposure (trifluridine, beryllium, arsenicals, petroleum products) cigarette smoking, vitamin A deficiency, and viruses like herpes simplex virus (HSV) type I.

Clinical Features

OSSN typically presents as a growth on the ocular surface and gives rise to symptoms like foreign body sensation, redness or irritation and rarely, diminution of vision due to high astigmatism or involvement of visual axis. It usually starts in interpalpebral conjunctiva and then grows and straddles the limbus and then may or may not involve the cornea. OSSN lesions mostly are slightly elevated and have a pearly grey appearance with tufts of vessels commonly known as sentinel vessels, with or without well-defined borders.

Morphological Classifiaction

Conjunctival Lesions

1. Gelatinous- Circumscribed gelatinous lesions are the most common. The nodular type is rapidly growing with a high incidence of metastasis to adjacent lymph nodes. The diffuse type is the least common and in the early stages presents as persistent redness of the conjunctiva,are slow growing and mimic chronic conjunctivitis. It is difficult to differentiate between benign and malignant lesions in these cases.
2. Leucoplakic- These are usually pre invasive.
3. Papiliform- These type typically are exophytic, strawberry like, with a stippled red appearance corresponding to its fibro vascular core. They are clinically benign.

Corneal Lesions

Corneal OSSN lesions are pre invasive, with a mottled ground glass sheet appearance which is opalescent. They have sharply defined fimbriated borders, the convex leading edge spreads in an arc away from the limbus and often white dots are present over the grey epithelium. They are usually avascular. These lesions are typically indolent, slow growing and prone to recurrence.

Differential Diagnosis

The differential diagnosis of OSSN includes :-

- Pannus
- Actinic disease
- Vitamin A deficiency
- Benign intraepithelial dyskeratosis
- Pinguecula
- Pterygium
- Pyogenic granuloma
- Keratoacanthoma
- Pseudoepitheliomatous hyperplasia
- Malignant melanoma and nevi

Diagnostic Tests

Exfoliative And Impression Cytology

Exfoliative cytology using a cytobrush is particularly suited as malignant cells have poor cell to cell adherence and tend to desquamate when located on the mucosal surface. Impression cytology using cellulose acetate paper (CAP) is as simple and inexpensive as exfoliative cytology with the added advantage of maintained cell-to-cell relationship.[18] However, CAP specimens require immediate processing. Impression cytology has been used widely as a non- invasive method for conjunctival biopsy for suspected OSSN. Using CAP for specimen collection, an 80% correlation was found between impression cytology, diagnosis and histopathology specimens obtained from incisional biopsy. Biopore membrane has better cell adherence and can be

stored for subsequent analysis making it the procedure of choice.[19] Within the intraepithelial group, keratinized dysplastic cells, often accompanied by hyperkeratosis, syncytial-like groupings, and nonkeratinized dysplastic cells are seen. Within the invasive group, cases with significant keratinization and an additional group of cases with little keratinization and sometimes also prominent macronucleoli are described. Keratinized cases are the most numerous in both the intraepithelial and invasive groups. It may also be used to monitor regression of lesion and response of the lesion to chemotherapeutic modulators. Mitomycin C (MMC) has gained acceptance for the treatment of OSSN especially in cases of recurrence or extensive disease where excision may jeopardise limbal stem cell function. McKelvie et al have followed patients after treatment with MMC for OSSN and, using impression cytology, demonstrated eradication of malignant cells, primarily by apoptosis, and a small amount of necrosis accompanied by inflammatory cells.[20] Normal cells undergo cytoplasmic enlargement and vacuolisation, and nuclear enlargement, but maintained a normal nuclear to cytoplasmic ratio. These changes persisted for a variable time following treatment, but resolved eventually. But using impression cytology as the only diagnostic tool has its pitfalls. Keratinising malignancies offer the highest chance of false negatives because of paucity of cells in the specimen and should be kept in mind in such cases. Cytological features that reliably differentiate carcinoma in situ (CIS) from invasive carcinoma are yet to be identified. Several patients may have histological CIN or partial thickness epithelial atypia adjacent to the invasive disease, which would not necessarily yield sheets of atypical cells if sampled by impression cytology. Endophytic lesions and orbital invasion cannot be identified with impression cytology, limiting its use as a diagnostic aid.

Histopathology

The specimens may be obtained from excision biopsies in small lesions which can be removed in toto or incisional biopsies in cases of large infiltrating lesions. Papillomas demonstrate papillary fibro vascular fronds covered by acanthotic epithelium. This epithelium may show varying degrees of dysplasia, however, the cells have normal polarity and the basal layers are often unremarkable. Preinvasive OSSN are classified as mild, moderate or severe depending on the degree of involvement of the dysplastic epithelium.

(i) Mild- CIN grade I: dysplasia confined to lower third of the epithelium.

(ii) Moderate-CIN grade II: dysplasia extends into the middle third.

(iii) Severe-CIN grade III: full thickness dysplasia, also called carcinoma-in-situ.

Invasive OSSN show nests of infiltrating cells that have penetrated the epithelial basement membrane and spread into the conjunctival stroma. These cells can either be well differentiated and easily recognized as squamous, or poorly differentiated and difficult to distinguish. The latter are more uncommon and more aggressive. Two types of cells may be seen interspersed with squamous cells in these tumours: spindle cells and mucoepidermoid cells.[21]

Electron Microscopy: Electron microscopy in cases of OSSN reveals excessive mitochondria, tonofilaments and endoplasmic reticulm; decreased desmosomes, alteration/ absence of basement membrane and deposition of fibrillogranular material between the basement membrane and bowmans layer.

Anterior Segment Optical Coherence Tomography (ASOCT)

In most of the OSSN, the diagnosis and treatment are formulated routinely using slit-lamp bio microscopy at 10 to 100 magnification in the clinical setting. In certain cases, biopsies are warranted for histopathology examination and diagnostic confirmation. Recent introduction of the optical coherence tomography (OCT) has provided ophthalmologists with a powerful tool for cross-sectional evaluation of various ocular conditions. The technology has undergone a few refinements, with transition from time-domain to spectral-domain OCT providing a better axial resolution and increasing scanning speed, leading to improved diagnostic imaging. A novel custom-built, ultra high-resolution, spectral- domain anterior segment OCT (UHR OCT) has been developed with an axial resolution of approximately 2μm for evaluation of corneal pathologic features.[22] The other commercially available time-domain OCT instruments have an axial resolution of 11 to 18 μm[23,24] making them insufficient for study of epithelial, Bowman's layer, or endothelial conditions. Therefore, the newly built UHR OCT provides a noncontact, non-invasive, and high axial resolution imaging for in vivo detection of various anterior segment processes. UHR OCT in ocular surface squamous neoplasia reveals epithelial thickening and increased reflectivity of the epithelium, and an abrupt demarcation from normal to abnormal tissue. Typically, there is sharp disparity in reflectivity of normal and diseased epithelium, allowing for exact localization of the tumor margins. The UHR OCT images of ocular surface squamous neoplasia may be helpful in the delineation of the tumor, and in the future may aid in the diagnosis as well as the ability to detect early subclinical recurrences.[25]

Confocal Microscopy

Ocular surface cytological examination with in vivo confocal microscopy is a simple, safe, and relatively non-invasive diagnostic tool that has been underused in the diagnosis and follow-up caring of patients with OSSN.[26–27] In vivo confocal microscopy could help with the initial clinical diagnosis of OSSN, estimation of recurrence, management of treatment, and evaluation of response to topical chemotherapeutic agents in patients with conjunctival and corneal squamous lesions. When compared with a subsequent biopsy, cytological evaluation with in vivo confocal microscopy was capable to distinguish different stages of OSSN in all the cases.[28] These findings were also

supported by other studies comparing the images from in vivo confocal microscopy with cytological samples obtained by either scraping or histopathological biopsy.[29,30] The new generations of confocal scanning laser microscopes have an axial resolution of approximately 4µm; however, the only disadvantage compared to UHR OCT is that they provide a transverse view without reference to neighbouring corneal layers.

Recent studies suggest that the accuracy of diagnosis with cytological in vivo confocal microscopy could be improved by the implementation of automated image analysis. This technique has been widely used in the interpretation of conjunctival and corneal exfoliative cytological analysis, and the acquired characteristics of cells were similar to those obtained from biopsies of OSSN.[31] Ocular surface cytological examinations, such as in vivo confocal microscopy, have several advantages in the diagnosis and management of OSSN. It is minimally invasive, almost painless, and can be performed in regular office settings. These features are useful when treating debilitated patients or patients who refuse to experience a more invasive surgery. Moreover, cytological image analysis can be transferred immediately to pathologists or ophthalmologists, elevating the efficiency of diagnosis.

In addition, in vivo confocal-microscopy sampling from various ocular surface regions can offer a real-time monitoring of the extent and condition of tumor, and guide further managements. However, comparison of concordance between pathologists and in vivo confocal microscopy with automated image analysis still deserves further investigation. Moreover, ocular surface in vivo confocal microscopic examination has its limitations. For instance, the maximum examining depth of this technology is 500 um, which impedes the detection of tumors inside the eye. In vivo confocal microscopic findings and the histopathological analysis showed high concordance in both the morphological features and the invasive extents of the tumors. Briefly, in vivo confocal-microscopy analysis can be reliable in predicting the grade of dysplasia that is based on cellular morphology, nuclear atypia, and nuclear to cytoplasmic ratio. The grading of OSSN also depends on the extent of the epithelial involvement that is better assessed by in vivo confocal microscopy compared with well oriented biopsy. In addition, cytological in vivo confocal microscopy assessment can differentiate between invasive and in situ tumors. A relatively accurate diagnosis and staging of carcinoma can be made from either histopathological evaluation of biopsies or cytological in vivo confocal microscopic assessment, although both techniques occasionally give false positive or false negative diagnosis. This study suggested that ocular surface cytological in vivo confocal microscopic examination was helpful in initial evaluation of subtypes of OSSN and could also be valuable in the setting of suspected recurrent tumours and in the follow-up evaluation of patients on topical chemotherapeutic agents. Although cytological in vivo confocal microscopy cannot replace biopsy, it can have an important role in the diagnosis and management of patients with OSSN in a non-invasive manner.[28]

Treatment

Surgery

Surgery has been the treatment of choice as a tissue diagnosis is considered essential before initiation of adjunctive therapy. Superficial excision (excision or incision biopsy) remains the important initial step in management as it is impossible to exclude invasive disease on clinical grounds or with impression cytology. Excision allows an immediate histopathological diagnosis, debulking, and excludes invasive carcinoma. The only disadvantage of primary excision alone is the high recurrence rate which ranges from 15% to 52%. Dissection of all abnormal tissue with a wide surgical conjunctival margin of 3 mm is usually sufficient. Conjunctival defect so created can be closed primarily (if less than three clock hours in diameter). Larger defects require either transpositonal conjunctival flaps, free conjunctival flaps from the other eye, or amniotic membrane grafts. Frozen section can be used to assess the adequacy of excision, and is accurate in delineating horizontal tumor spread. Bunns modification of Moh's technique of tumor margin surveillance may also be used. In this the free conjunctival edges are excised by 2 mm if residual tumor is evident even after excision of a 2 mm surgical margin.[32] Enucleation, and rarely exenteration may be required in cases of intraocular or intraorbital spread. In all cases a no touch technique is used, and direct manipulation of the tumour is avoided to prevent tumour seeding.

Cryotherapy

This modality is often used in conjunction with surgery. It acts by directly destroying the tumor cells by lowering the temperature and also by causing ischemic necrosis. Intraoperative cryotherapy is commonly used as adjunctive therapy as it is known to decrease the recurrence rate by destruction of any residual tumour tissue beyond the horizontal or deep surgical margin of the wound. It has the advantage of reaching both tumor cell islands and deeply infiltrated cells,thus obviating the need for radical surgery.

A nitrous oxide cryoprobe tip (2.5 or 5mm) is used to form an ice ball extending 2mm for conjunctiva,1mm for episcleral tissue and 0.5mm for the cornea. A slow duration freeze with a slow thaw, repeated two times (freeze-thaw-refreeze) is recommended. It is important to include the limbal region during cryotherapy, and not apply the cryoprobe for more than three seconds. Both extensive surgical excision and cryotherapy can cause limbal stem cell insufficiency.

Radiotherapy

Various sources such as strontium-90 (beta irradiation) and radium (gamma radiation) were used earlier. But given the high incidence of side effects and prolonged duration of treatment required, it is rarely used now.

Chemotherapy

Topical chemotherapy is inexpensive, simple and reduces the risk of limbal stem cell deficiency, and obviates the need for clear tumor margins by treating the entire ocular surface, including the potentially dysplastic cells.[33] However, the obvious limitation is the limited drug penetration in larger tumours, and a possibly deleterious effect on the ocular surface and nasopharyngeal epithelium on prolonged use.

1. **Mitomycin C**: It is an anti-tumour antibiotic that preferentially inhibits DNA synthesis in the G1 and S phases. As the hypoxia required for the intracellular reduction of MMC is greater in tumour tissue, it exhibits a certain degree of selectivity. MMC appears to produce cell death in OSSN by apoptosis and necrosis.[34] MMC related changes may persist in ocular surface epithelium for at least 8 months following MMC therapy. It is used in the concentration of 0.02-0.04% four times a day with one week on and one week off in alternating cycles for a maximum of 8 weeks. The one week on, one week off regimen prevents damage to more slowly dividing epithelial cells and limbal stem cells, allowing them to repair their DNA. Allowing time for complete epithelial healing before application of MMC is important in avoiding the more serious complications such as corneal epitheliopathy, scleral ulceration, uveitis, cataract, and glaucoma.
2. **5 Flourouracil**: It is an antimetabolite that acts specifically during the S phase of the cell cycle. It is converted to 5-F DUMP, which inhibits thymydilate kinase thus preventing DNA and RNA synthesis. Both MMC and 5FU are currently being used four times daily for 1-2 weeks in a pulsed fashion, the treatment being repeated after every 1-2 weeks. This one week on and one week off drug regimen has the added advantage of good efficacy and better tolerance.

Immunotherapy

Interferon alpha2b (INF-α2b) is a naturally occurring glycoprotein which binds to cell surface receptors affecting intracellular events resulting in anti tumor and antiviral properties. It has been used in the treatment of many disorders, including hepatitis[35], cervical intraepithelial neoplasia[36] and cutaneous squamous cell carcinoma.[37] Its efficacy in treatment of OSSN may be explained by the oncogenic link between HPV and OSSN. Topical drops and subconjunctival injections of interferon-α2b (INF-α2b) have been used as off-label therapy to treat OSSN. It has been used for extensive, residual, recalcitrant, multifocal or diffuse lesions and for those that involve the visual axis where surgery is not the treatment of choice. Interferon alfa2b is an important treatment modality for recalcitrant OSSN, effective in both, primary tumours unresponsive to treatment, as well as recurrences. It is more toxic than MMC and it usually takes a longer duration for complete resolution compared to MMC, so is not started as a first line of therapy and preserved for lesions non responsive to topical MMC. IFN-albha 2b drops 1 million international unit/ml (IU/ ml) is used four times a day until resolution, and a month thereafter. Subconjunctival injections 3 million IU/ml have also been used. Median time for resolution has been reported as 54 days (range 28-188 days),with a mean follow up ranging from 2.9 to 18 months.[38] Medical therapy with Interferons has the advantage of treating microscopic disease that may be present throughout the entire ocular surface.

No study has specifically evaluated the optimal dosage of IFNα2b therapy. The two doses recommended are 1 and 3 million IU/ml. No data exist on whether the higher dose is justified with respect to time to lesion resolution, disease recurrence and side-effect profile. Hence the two standard doses - 1 million and 3 million IU/ml were retrospectively compared for efficacy in treatment of OSSN. The 1 million IU/ml dose was the empirical dose used in the initial study. The mean time to tumour resolution was about 12 weeks. The 3 million IU/ml dose was later compounded in an attempt to shorten the duration of treatment. Although the study was small and therefore had limited power, the findings suggested that no large clinical differences exist in tumour response between the two IFN-α2b doses. There may be a trend for faster resolution and increased side effects in the 3 million IU/ml group. As with all retrospective studies, the conclusions must be interpreted cautiously due to the limitations of the study. The conclusions are based on the assumption that the baseline characteristics between the two groups were similar. The groups were not identical with respect to many factors. This may have skewed the results if the CIN lesions in the 3 million IU/ml group were more aggressive than in the 1 million IU/ml group.

Patients with both primary and recurrent disease were included in the analysis. Although an equal proportion of patients in each group had recurrent disease, this may have also skewed the results. In addition, more patients with bilateral disease were treated with 3 million IU/ml of interferon. This introduces a correlation bias as eyes of the same patient are expected to respond similarly to treatment. Finally, there was inclusion of both biopsy-proven and clinically diagnosed lesions in the analysis; however, there was justification in this decision due to the high clinical accuracy of diagnosing CIN. Due to the small number of eyes with CIN treated with topical interferon therapy alone, and the retrospective nature of the study, these limitations are difficult to eliminate. In light of these overall considerations, the recommendation was to continue using the 1 million IU/ml dose in the treatment of CIN as before.[38]

Pegylated Interferon Alpha 2b

Pegylation of therapeutic proteins is a well-established method for delaying clearance and reducing immunogenicity.[39] Pegylation has been used to modify a variety of proteins, including tumor necrosis factor-

alpha (TNF-α) and human growth hormone.[40,41,42] In some cases, the pegylated protein has been more effective than the native protein.[29,40] Pegylated interferon alpha 2b (PEGIFNα2b, PEG Intron, Schering- Plough, Kenilworth, NJ) is a derivative of recombinant interferon alpha 2b and was developed to reduce the clearance of traditional recombinant interferon alpha 2b. Attachment of a single straight-chain polyethylene glycol (PEG; molecular weight of 12,000 Da) moiety to interferon alpha 2b does not alter the volume of distribution or spectrum of activity, but does significantly decrease renal clearance, increasing plasma half-life ten fold compared to non-pegylated interferon.[43,44] This has allowed the dosing of the pegylated interferon to be once weekly for systemic diseases as compared to non-pegylated interferon (recombinant), which is dosed 3 times per week to daily. The safety and efficacy of PEGIFNα2b has been established in patients with chronic hepatitis C, renal cell carcinoma, and chronic myelogenous leukemia (CML).[45] A recent randomized controlled study comparing quality of life and toxicity of PEGIFNα2b to non-pegylated interferon found less systemic side effects in the PEGIFNα2b group.[46] One potential disadvantage of the PEGIFNα2b over non-pegylated interferon is its much higher cost.

A study was conducted to evaluate the effectiveness and side-effect profile of this modality in OSSN.[47] It was found that PEGIFNα2b was effective in this small pilot study for treating OSSN as complete clinical resolution of the lesion was seen in all patients. A mean of 3 injections were needed for tumor resolution in a case series using PEGIFNα2b. In comparison, a previous case series using subconjunctival and topical recombinant interferon for the treatment of CIN reported that a mean of 5 injections were needed for tumor resolution.[48] Pegylated interferon costs approximately 3 times as much as recombinant interferon. Due to the small number of patient in this study, it cannot be commented whether the extra cost of PEGIFNα2b is justified in the treatment of OSSN. Future studies will be needed to evaluate the safety, efficacy, side effects and long-term success of PEGIFNα2b for the treatment of OSSN and to determine whether it is preferable over recombinant interferon given the significant cost differential between the two medications.

Recurrence

Recurrence rates of OSSN ranges from 15-52%, average reported being 30%. Recurrences are higher in case of inadequate excision margins, and occur usually within two years of surgery. These typically exhibit a more aggressive behaviour because of the tissue disruption associated with the primary excision theoretically enhancing the ability of the tumor cells to enter the eye. The main predictors for recurrence include age, histological grade of the lesion, adequacy of margins at initial excision, corneal location, larger size (>2 mm), and a high proliferation index. Immunostaining with antibody to Ki-67, which is a nuclear antigen expressed in proliferating cells, allows evaluation of the growth fraction of normal and neoplastic cells yielding the proliferation index.[49]

Recommended Therapeutic Strategy and Current Therapeutic Practice

The recent advances and the current status of the diagnostic modalities and management of squamous neoplasms have been reviewed by Basti et al[50] and have made the following recommendations. Although the clinical diagnosis of in situ disease is high (86%), invasive carcinoma is much less often recognised (35%). Larger lesions and those with hyperkeratosis are more likely to be correctly diagnosed preoperatively. Impression cytology does not reliably distinguish in situ from minimally invasive disease, and therefore has limitations in the accurate diagnosis of OSSN.

1. **Suspected OSSN 1-3 clock hours** :- Complete excision biopsy is recommended

- If residual tumor is there in margins, chemotherapy with MMC is given, 3 monthly review is done to evaluate tumor resolution. Thereafter, follow up every six months
- If tumor margins are free of tumor, 3 monthly follow up done for a year to confirm absence of recurrences; thereafter follow up every six months.

2. **Suspected OSSN 3-6 clock hours** :- Biopsy done to evaluate invasiveness of tumour

- If preinvasive: start chemotherapy with MMC
  - Monthly follow up, with quarterly evaluation for tumor resolution. If complete resolution, follow up every six months
- If invasive: start topical chemotherapy to achieve chemo reduction
  - Surgical excision of any residual tumor with cryotherapy to bed. Cover exposed area with amniotic membrane graft.
  - Monthly follow up, with quarterly evaluation to confirm absence of tumor recurrence. Thereafter, follow up every three months.

3. **Suspected OSSN >6 clock hours :-** Biopsy to decide if invasive or preinvasive

- If preinvasive: start chemotherapy
  - Monthly follow up, with quarterly evaluation of tumor resolution. If complete resolution, follow up every six months
- If invasive: high dose chemotherapy with MMC
  - If complete resolution, monthly follow up for a year, quarterly thereafter.
  - Partial resolution, chemoreduction achieved, surgical excision of any residual tumor and cryotherapy to bed. Cover exposed area with amniotic membrane graft.

- Monthly follow up, with quarterly evaluation to confirm absence of tumor recurrence. Thereafter, follow up every three months
- If >6 clock hours after chemotherapy, palliative treatment with radiotherapy.

In a survey of ophthalmologists treating OSSN, almost 54% ophthalmic surgeons believed that sufficient evidence exists to justify the use of mitomycin C in the treatment of OSSN, and 15% felt that the published literature justified the use of 5-fluorouracil or interferon. About 50% of ophthalmic surgeons always performed a biopsy before institution of topical therapy. The reported use of topical chemotherapy as an adjunct to surgical excision increased with the size of the lesion. Nearly 45% of the respondents utilized topical therapy along with surgery for lesions greater than 8 mm in diameter.[51]

Conclusion

Squamous lesions of the cornea and conjunctiva are uncommon but demand appropriate attention due to the potential for visual loss and systemic morbidity and mortality. Newer modalities of diagnosis allow non invasive evaluation which correlates well with histopathological tissue diagnosis. Further refinements of modern therapeutic options will allow cell specific anti-cancer treatment of these lesions with preservation of the limbal stem cells and ocular surface.

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