CERVICO-FACIAL AND INTRA-ORAL ACTINOMYCOSIS

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THESIS SUBMITTED FOR THE DEGREE

OF MASTER OF DENTAL SURGERY

DEPARTMENT OF DENTAL SURGERY

GLASGOW DENTAL HOSPITAL AND SCHOOL

UNIVERSITY OF GLASGOW 1976

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ACKNOWLEDGEMENTS

As John Donne, the sixteenth century poet and philosopher said; "No man is an island, entire of itself", so am I conscious of the help and support of so many in the Glasgow Dental Hospital and School in the preparation of this thesis. Co-operation and advice has been freely given not only from the Dental Surgery Department but also from the Oral Pathology and Oral Microbiology Departments.

In my own department, I should like to thank all my colleagues for their support, and in particular Professor J.C. MacDougall and Mr. H.D. Campbell who have shown sustained interest in the work.
Dr. D.G. MacDonald, Senior Lecturer in Oral Pathology, and Mr. W. Marshal, Senior Chief Medical Laboratory Technician in that department have been constant enthusiasts and advisors and I am grateful to them both.
I have spent much time in the Oral Microbiology Unit where Mr. T.W. MacFarlane, Lecturer in Charge has been invaluable in his assistance and counsel. I also owe a great debt to Mr. D. McKenzie, Senior Medical Laboratory Technician, who bore the brunt of laboratory innovation during the development of the sections involving the fluorescent work, and who was unstinting in his efforts to help me.

The fluorescent antibody conjugates were generously provided to me by Dr. Morris T. Suggs, Director of the Biological Products Division of the

Centre for Disease Control, Atlanta, Georgia, to whom I would take this opportunity to thank most sincerely.

For the study on denture wearers, I was ably assisted by Mr. I.B. Watson of the Prosthetics Department, and thanks are also due to Mr. J.B. Davies and staff for photographic work, and Mrs. M. Pettifor and Mrs. R. Steen for secretarial work.

Finally I am indebted to the University of Glasgow and Professor T.C. White, Director of Dental Education for the opportunity to carry out this study.

PREFACE

Actinomycosis is an infective non-contagious disease characterised by a granulomatous reaction in the host tissue. It is most commonly found in the cervico-facial region related to the jaws and in particular the mandible. It is, however, essentially a soft tissue infection with direct involvement of bone a rarity. Other sites of the disease are lung and abdomen, with skin only infrequently involved.

The infection is caused by Actinomyces Israelii, an anaerobic micro-organism which exists as a commensal in the oral and pharyngeal regions and which, if given access to a suitable oxygen-free environment, may assume a pathogenic role. Cervico-facial actinomycosis although relatively uncommon is of importance to dental surgeons, since the disease is frequently related to an infected tooth or oral wound, and the patient often presents first to the dentist for advice.

During my professional career, several patients with cervico-facial and intra-oral actinomycosis have undergone treatment in Glasgow Dental Hospital and School, and I have been fortunate in being involved in the management of the majority of them. The present work was stimulated by these cases which proved diverse in their presentation and which required individual assessment in formulating treatment plans.

Aims and Objectives

Professor Jennett writing in the Journal of the Royal College of Surgeons of Edinburgh of January 1974 in an article entitled "Surgeons of the Seventies" noted the general underestimation of the value of the retrospective survey and the fashion today to discredit any research other than the planned prospective study. It is, therefore, without apology that much of this work is of a retrospective nature.

The thesis is divided into two sections.

Part I Concerns information gained from the clinical courses of the accumulated cases, and an attempt is made to relate the information so gathered to the current concepts of the disease which have been published, and where possible to add to this knowledge.

Part II Concerns both retrospective research and prospective studies. In this section, an attempt is made to understand the possible pathogenesis of actinomycosis, and conclusions are reached which may influence clinical treatment and lead to a clearer understanding of actinomycotic infection in the oro-facial region.

Two appendices are included which consist of abbreviated case reports of the cervico-facial cases and intra-oral cases respectively. The former are numbered 1 to 15

and the latter A to L. These cases are referred to in the text by such annotation, and the tables also correspond directly to the appendices.

Publications

1. Low Grade Osteomyelitis of the Jaws with Actinomycosis. (1974)
   International Journal of Oral Surgery 3: 60-64.
   (with D.G. MacDonald)
2. Cervico-facial Actinomycosis : Aetiology, Diagnosis and Treatment. (1974)
   Edinburgh Dental Hospital Gazette 13: No.3.
   (with T.W. MacFarlane)
3. Intra-oral Actinomycosis. Report of five cases (1975)
   Oral Surgery Oral Medicine Oral Pathology, 39: 547-552.
4. Cervico-facial and Intra-oral Actinomycosis: A 5 Year Retrospective Study. (1975)
   British Journal of Oral Surgery, 13: 172-182.
   (with D.G. MacDonald and T.W. MacFarlane)

SUMMARY

Actinomycosis was first recognized as a specific disease late in the 19th century. Confusion as to the nature of the infecting micro-organism and the mode of its entry into soft tissue led to two diverse theories on the pathogenesis. On the one hand it was believed that the microbe was an aerobic species which grew on agricultural land and which was introduced to the body from the habit of grass or straw chewing, whilst on the other hand the organism was held to exist as a commensal in the oral cavity which could under certain circumstances become an active pathogen. Many investigators contributed to these basic arguments, but the overwhelming weight of evidence came down on the side of the endogenous theory. Despite this, many texts still perpetuate this misconception.

Cervico-facial actinomycosis is the most common site of the disease, and it is most prevalent in the second and third decades affecting males more frequently than females in a ratio of approximately 2 to 1. There is a close asscciation between the teeth and cervico-facial actinomycosis. The vast majority of cases are related either to an apical dental abscess or follow some form of oral trauma, usually an extraction. Two clinical variants of the disease can be recognized. The acute infection is often related to a septic tooth and is shown to affect a younger age group, whereas the chronic infection more often is a sequel to an extraction

and affects an older age group. In both groups, the lower molar region is predominantly involved.

The diagnosis of actinomycosis involves a combination of astute clinical awareness and microbiological technique. The use of specific fluorescent antiserum in the diagnosis of cervico-facial actinomycosis is shown to be of value in the early recognition of the disease, thus allowing the clinician to formulate and implement correct therapeutic drug regimes at an early stage. The value of bacteriological examination of intra-oral specimens is highlighted as is the limitation of diagnosis from formalin fixed material.

Treatment of cervico-facial actinomycosis involves a combination of surgery and antibiotic therapy, normally penicillin. The difficulty of laying down rules on the duration of therapy is clear, since each case requires individual assessment. The host reaction to actinomycotic infection is a fibrosis which may lead to scar formation on resolution.

Investigations into the occurrence of actinomyces in relation to bony sequestra may be an indication of the mechanism by which the organisms gain access to the soft tissues, and the frequency with which the lower molar region is implicated. Although actinomycosis in bone is rare, the sequestrum may provide a suitable environment for these bacteria to thrive before invading the soft tissues which have been opened to the infection by the fracture of the buccal or lingual alveolar bone which

originally provided the sequestrum.

Logically, the bacterial population of the dental plaque in the lower molar area must be considered and investigations showed that A. israelii is present in larger numbers in this area than in the anterior region of the mouth. This study also indicated that A. israelii is more prevalent than A. naeslundii per unit volume of plaque in both anterior and posterior regions and in total numbers. A. naeslundii showed no significant difference in numbers anteriorly compared with posteriorly.

Studies on the flora obtained from the fitting surfaces of upper dentures in edentulous subjects confirmed the close relationship between actinomyces and the natural dentition. Finally, it was shown that in vitro, polymorphonuclear leucocytes were able to ingest both cocco-bacillary and filamentous forms of actinomyces. The impediment to more rapid resolution of the infection would hence appear to lie elsewhere. Excessive fibrous walling off by the host tissue does offer one explanation for this slow healing response particularly in the more chronic form of the disease. Furthermore, the "sulphur granule" itself may act as an effective barrier to successful phagocytosis.

Actinomycosis in the oro-facial region remains a fascinating disease to the medical and dental professions. It is one of the few pyogenic infections in the area which can be attributed to a specific bacterium. Most abscesses of the jaws are found to be due to a mixture of pathogens,

and actinomycosis is no exception. The isolation of actinomyces, however, forewarns the clinician of the likelihood of persistence and a marked fibrotic reaction from the host tissues. Its early diagnosis is of paramount importance to treatment, and although the disease is comparatively rare, in the words of W.D. Miller written as long ago as 1890 "We consequently have here again another dangerous source of infection in the human mouth, which the dentist and physician do well not to lose sight of."

Chapter 1

Historical and Aetiological Review

Early History

Earliest reports of actinomycosis were made in the latter quarter of the 19th Century, and, although much of the work was of the highest quality in terms of description, confusion occurred in the differentiation between human and animal disease, and also in the aetiological micro-organisms. Unfortunately, much of this confusion still persists although the facts are now unequivocal.

The name actinomyces or "Ray fungus" was introduced in 1877 by Bollinger who isolated a micro-organism from purulent exudate from a cow with "lumpy jaw." In co-operation with Harz, a botanist, he described the micro-organisms forming a mycelial mass with peripheral radiations and this was the first accurate report of the disease in cattle and of the aetiological bacterium. Bollinger named the micro-organism actinomyces bovis. Earlier descriptions of disease or bacteria are to be found but with less precision and hence of more doubtful accuracy. Amongst these was a description by Cohn in 1875 of a streptothrix which he isolated, but did not culture, from a human tear duct, and which bore a marked resemblance to the actinomyces described by Bollinger and Harz. Earlier still, Von Langenbeck in 1845 is known to have described a "fungus disease" in man which on reflection was probably actinomycosis.

However, it was between 1878 and 1891 that the workers most identified with actinomycosis in man came to the fore, namely Boestrom and James Israel and Max Wolff. The argument between Boestrom on the one hand and Israel and Wolff was fundamental and concerned the nature of the infecting agent, and its access to the tissues. The respective arguments were known as the exogenous (Boestrom) and the endogenous (Wolff, Israel) modes of infection, and it is worthwhile considering these in more detail, since the confusion which occurred at that time still persists.

Boestrom in his publication in 1890 reported bacteriological investigations of both animal and human origin. His contention was that the infecting micro-organism was an aerobic species although he had isolated both aerobic and anaerobic "fungi" from his cases. He concluded that the aerobic fungus was the infecting agent but was singularly unsuccessful in his attempts to induce the disease in animals using his aerobic culture. He claimed that the infection arose from fungi outwith the body which were found in abundance in soil and vegetable matter. It was from this time that the misconception of a relationship between agrarian workers and of grass and straw chewing habits and the disease took origin.

In 1891 Professor Wolff and James Israel published their investigations into the bacteriology

of actinomycosis in man. In this paper, they contrasted the essential differences between their findings and those of Boestrom, and selected four basic points of difference between the results. These differences were:-

1. The relationship between the microscopic culture appearance and rate of growth of the micro-organism.
2. The energy of the growth both aerobically and anaerobically.
3. The microscopic appearance of the fungus during the early culture period.
4. Disease production in animals using pure cultures of the bacterium.

Much stress, however, was placed on animal experiments in which Wolff and Israel had successfully infected with a human actinomyces culture 22 out of 23 animals. In all these animals, lesions were produced which on microscopic examination revealed the typical actinomycotic colonies. In 10 cases they had cultured the needle shaped organisms back onto artificial media, thus fulfilling Koch's criteria for infective agents, and hence they indicated their fundamental disagreement with Boestrom who had had "uniformly negative" attempts to produce the disease in animals.

In a publication in 1885, Israel had collected 38 cases and had attributed actinomycotic infection of the lower jaw to colonisation of the ray fungus in

carious teeth. He was thus in no doubt as to the origin of the infection though Ponfick in 1879 had proposed any minor injury or defect of the mucous membrane as a possible point of entry. The two theories were linked by the common need for local injury to allow the fungus access to the tissues.

Endogenous Mode of Infection

In the following years evidence favoured the Wolff-Israel theory of endogenous infection.

Homer Wright in 1905 was prominent amongst those who supported Wolff and Israel in their contention that Boestrom had erroneously linked his aerobic organism with actinomycosis. In his comprehensive review of the literature and personal experience with 13 human cases and 2 bovine cases, he compared and contrasted the two theories. He objected to Boestrom's arguments on two main points. Firstly that "micro-organisms had been found in only a very small proportion of his cultures and in no larger proportion than it would be possible for the micro-organism he describes to have appeared as a contamination." To explain this objection, it is necessary to understand that Boestrom had obtained pus from 11 cases of actinomycosis in cattle. In all, around 700 culture tubes were innoculated, and although he obtained his aerobic growth from 7 of the 11 cases, only 12 of the 700 culture tubes were positive. These 12 positive isolates grew luxuriantly on sub-culture

and Wright was unwilling to accept that such a free-growing micro-organism would not have been noted in a far higher proportion of his primary cultures. His second main objection was a re-iteration of Wolff's - namely that Boestrom had failed to produce typical actinomycotic lesions in experimental animals. Wright compared his own results with those of Wolff and Israel with regard to both cultural characteristics and animal innoculation. He concluded that the actinomyces of Wolff and Israel and that which he had isolated were one and the same species.

In his general concluding remarks, Wright stated that he considered Actinomyces bovis a "normal inhabitant of the secretions of the buccal cavity and of the gastro-intestinal tract, both of man and animals, but I have no proof of this to offer at the present time."

In 1910, Lord demonstrated organisms having similar morphology and staining reaction to actinomyces in 11 smears and from 5 sections of carious teeth. He further investigated by innoculating guinea pigs with material taken from carious teeth and tonsillar crypts and produced omental tumours histologically identical with actinomycosis in 60% of cases. His theory on the relative rarity of human actinomycosis was that pyogenic bacteria, by their more rapid multiplication, did not permit of the actinomyces "taking root" in the tissue. Colebrook in 1920 also favoured the dental source of infection in his study of 28 cases.

Naeslund in 1925 described three groups of actinomyces isolated from the human mouth. These were Group A, preferably aerobes; Group B, preferably anaerobes and Group C which grew aerobically and anaerobically. The first group he equated with Boestrom's aerobic cultures which he claimed were not indigenous to the oral cavity but were "chance organisms which have invaded the mouth from outside." His anaerobic group he claimed, were identical culturally with the preferably anaerobic forms cultivated from actinomycotic infection and which were commonly referred to as Wolff-Israel's actinomyces. Lord and Trevett in 1936 discussed the pathogenesis of actinomycosis and considered the aetiological relationship of the organisms of Wolff and Israel to actinomycosis as established, and held that Naeslund has proved that these organisms were harboured in the normal mouth.

Sullivan and Goldsworthy in 1940 attempted to relate the anaerobic strains of actinomyces from clinically normal mouths to those from actinomycotic lesions. They isolated 5 strains from 100 periodontal pockets and 1 strain from 24 carious lesions. With these 6 strains, they carried out studies of cultural characteristics and compared the biochemical sugar fermentation reactions from them with 6 strains from proven actinomycotic lesions. Their results suggested that the two groups were identical, and furthered the

idea that dental foci existed for the actinomyces. Sullivan and Goldsworthy were essentially working in dental clinics and they had noted the relationship between cervico-facial actinomycosis and oral trauma. They cited Warwick's case reported in 1923 of a carious tooth being found in the lung of a patient with pulmonary actinomycosis, and indicated that Israel's theory of the colonisation of carious teeth by the ray fungus was hence even at that early stage an astute observation. Later papers were to show the typical actinomycotic colonies associated with periapical dental lesions. (videinfra). The work of Erikson (1940) clarified the essential differences between the animal and human infection by clearly demonstrating the two distinct infecting micro-organisms. She proposed that the human infecting agent be known as A. israelii and that of the animal infection A. bovis. Confirmation of this work came from Thompson (1950) who, however, did indicate that isolated cases could occur of human disease from A. bovis and vice versa. Thompson and Lovestedt (1951) proposed the name Actinomyces naeslundii for the facultative anaerobic species first described by Naeslund in 1925, and maintained that this organism was saprophytic and was frequently mistaken for A. israelii in the mouth.

The Agrarian Link

The problem of the exogenous mode of entry was tackled by some investigators in another way. Having

been influenced by the Boestrom arguments, several clinical surveys looked for possible links between their patients and "vegetable products." Hence Acland's series (1906) of 109 British cases contained 17 who either chewed straw habitually or were of agricultural background. Foulerton in 1913 collected 78 cases and noted that 24 were in occupations bringing them into close contact with vegetable matter. His link was however rather tenuous, and such occupations as tailor and footman were considered relevant. In one of his cases, a medical student, he had ascribed her infection as having been brought about by walking through fields and chewing grass while on holiday. Cope in 1915 reported a case where a 10 year old child had developed actinomycosis after swallowing a blade of grass which had lodged in the tonsil, and Mattson in 1922 claimed that of 26 cases, 11 admitted to a straw or grass chewing habit. In a later survey in 1951, Porter reported on 91 cases in the North East of Scotland of whom 20 were farmers and concluded that agricultural workers were more liable to the disease than others in the population. This was probably a more pertinent survey since he had submitted his figures to statistical analysis. Sanford Magath in 1921 also supported the agrarian connection in their survey in the Mayo Clinic. 57 out of 90 of these cases had agricultural connections. However in this same paper, they had collected 119 cases from

various sources in which only 16 were farmers, and Davis in 1941 in his study of 46 cases recorded that only 3 had admitted to a grass chewing habit. Holmes (1958) and Bramley and Orton (1960) rejected the theory with only 1 case remotely connected in 25 cases. Direct contagion from animals was advanced by Mattson (1922), but generally this was not a popular theory. To come to a conclusion therefore is difficult. Grass chewing can almost certainly be dismissed as providing a source of the infecting organism though it may cause injury and hence allow access to the endogenous micro-organisms. Agrarian workers would appear from some surveys to be more liable to the infection but as Lesney and Traeger (1959) indicated, this concept was ingrained for so many years that this connection was sought perhaps too avidly giving rise to erroneous conclusions.

Induction of Animal Disease

The production of disease in animals by innoculating bacterial preparations was of great importance in the aetiological argument. Most researchers had noted difficulty in causing progressive disease in animals using the anaerobic actinomyces. Wolff and Israel were able to produce actinomycotic colonies in their animals - mainly rabbits - but there did not appear to be any progression of the disease, merely a characteristic nodular reaction of the tissues around the area. Wright (1905) also induced disease in animals but noted the lack of extension of the disease process within the

animal. Indeed he stated that very few of the cases showed active multiplication of the bacteria within the animal body. However Mathieson et al in 1935 in a paper entitled "Allergic reactions of actinomycetes" produced progressive fatal actinomycosis by repeatedly sensitizing guinea pigs to innoculations with actinomyces cultures. They suggested that repeated exposure of the host to the bacterium led to sensitization of the animal and progressive fulminating disease. Slack in 1942 used this repeated innoculation technique with his animals, and was successful in producing widespread extension of the disease culminating in death. There are grounds, therefore, for believing that humans may also be sensitized to the bacterium before active disease supervenes, though this contention would be extremely difficult to prove. Nevertheless the oral cavity is subjected to repeated trauma, and it would be possible for such a sensitization to occur.

Mixed Infection

Actinomycosis is widely recognized as being primarily due to the anaerobic actinomyces israelii, but from first investigations, most cultures from active cases have been mixed infections - with greater or lesser predominance of the actinomyces themselves. Wright (1905) thought that these accompanying bacteria were important in the extension of the disease but considered that the actinomyces bovis was capable of

acting as the sole infecting agent. In 1912 Klinger described a small Gram negative bacillus which he noted in the actinomycotic pus. Colebrook in 1920 indicated that this concomitant micro-organism which he called bacillus actinomycetem comitans was noted in 80% of his 28 cases, and Per Holm refers to this mixed infection in a most comprehensive review in 1950. Per Holm studied the pus from no fewer than 650 patients with closed actinomycotic lesions in Denmark over a 15 year period. He did not find a single case in which actinomyces were the sole micro-organisms. He found the concomitant bacterium to be various Gram negative bacilli including the B. actinomycetem comitans of Klinger, diverse anaerobic streptococci and several others, and he maintained that actinomycosis was a combined infection with anaerobic microbes of the actinomyces species acting synergistically with other various bacteria. The following year (1951) he reported 9 cases in which penicillin had been unsuccessful in eradicating the infection. In two cases in particular - one of the cervico-facial region and the other of thorax - he noted that the actinomyces species was eliminated by penicillin therapy but the patients remained ill. Culture of the discharge revealed B. actinomycetem comitans, and he concluded that these other microbes had a certain conditional or relative virulence.

Trauma

The question of trauma as a predisposing factor has long been recognized, and is certainly relevant in the pathogenesis of actinomycosis. It is clear that the actinomyces israelii is a commensal organism in the oral cavity which under certain circumstances can become pathogenic. Two main theories exist to explain the possible mode of access to the deeper tissues of the jaws. The first is the carious tooth which forms a periapical abscess and may then present as a facial swelling usually in the submandibular region. In this form, the abscess may be clinically indistinguishable from the acute pyogenic dental abscess. There can be no doubt that this form of access exists, as actinomyces have been noted in pulps (Hardwick and Newmann, 1955; Villa, 1957) in periapical granulomata (Browne and O'Riordan, 1966; Hamner and Schaefer, 1965; Kalnins, 1971; Gee and Sullivan, 1940; Kapsimalis et al, 1968; Gold and Doyne, 1952; August and Levy, 1973; Goldstein et al, 1972) and in apical dental cysts and dentigerous cysts (Sprague and Schaefer, 1963; Martinelli and Rulli, 1967). As has been noted Israel was the first to attribute infections of the lower jaw and neck to colonisation of "ray fungus" in carious teeth, and most of the other principal investigators also noted this possible route. In these cases, the infection is related to a clinically non-vital

infected tooth. Danielewiczowa et al (in Brommer & Brommer, 1971) in a report of 260 cases in Poland between 1952 and 1969 reported that of 121 cases where local factors could be implicated in the occurrence of cervico-facial actinomycosis, no fewer than 87 were related to teeth with gangrenous pulps. Mitchell (1966) found 23 of his 46 cases were related to infected teeth, and many other reports including those of Rud (1967), Holmes (1958) Norman (1970) include cases which were directly related to infected teeth.

The other method by which the organisms gain access to the deeper tissues is through an oral wound, most commonly a dental extraction or jaw fracture. The prevalence of lower molar teeth in both categories is striking and will be further considered at a later stage. The penetration through the lower alimentary tract by the organisms is less easy to understand. Cope (1938) cited cases following perforation of a peptic ulcer and through ruptured appendices. Pulmonary or thoracic involvement may be subsequent to aspiration or escape of the organism into the mediastinum through the oesophagus. Accidental inhalation of carious tooth fragments, Cope noted, had led to actinomycotic abscesses in lung with the dental tissue forming the central core (Warwick 1923). In 1973 Brown, in a review of 181 cases in the Armed Forces Institute of

America, cited previous surgery or chronic lung disease as possible predisposing factors in abdominal and pulmonary actinomycosis but was emphatic in stating that the majority (100 cases) has no such history.

Further discussion of the role of trauma will be confined to cervico-facial actinomycosis. A large number of cases in the form of case reports have been published, and it is apparent that extraction of teeth or fracture of the mandible is commonly found. Goldsworthy in 1947 considered cervico-facial actinomycosis as a disease of the "age of exodontia" while Sanford and Magath (1921) found that in most of their cervico-facial cases, there was a history of preceding "tooth or tonsil trouble." Porter (1951) reported that of his 72 cervico-facial cases, 3 had previous "injury" to the area and 12 had a definite history of extraction. Rud in 1967 reported 4 out of his 10 cases were related to a socket while Holmes in 1958 reported that half of his 12 cases had an unequivocal relationship to dental extractions. In a most comprehensive clinical review of 46 cases, Mitchell in 1966 divided his cases into post traumatic and those with no history of physical injury. 50% of these cases were related to oral injury, 21 following exodontia and 2 consequent to fractured mandible. In a Canadian review in 1942, Hamilton reported

15 of 25 mandibular cases related to extraction. Examples of cases following fractured mandible are those reported by Gruber (1952) Hylton et al (1970) Ciebien et al (1964) O'Mahoney (1966) and Jurgens (1962). Tokiwa et al (1959), however, reported 283 cervico-facial cases from Tokyo Medical and Dental University of which only 16.1% were related to extraction. They concluded that as far as the aetiology was concerned, Japan was at variance with most European and American reports.

Chapter 2

Epidemiology and Occurrence of Actinomycosis

Actinomycosis occurs world wide and involves all races. Brown 1973 in a study of cases filed in the Armed Forces Institute of Pathology recorded Caucasian, Negro and Mongoloid patients, and although most originated in the United States he noted cases from Europe, Africa, Asia, South America and Australia. Many attempts have been made to evaluate the prevalence of the disease (Porter 1951, Cope 1930, Sanford & Magath 1921, Acland 1906, Foulerton 1913) in terms of an annual mean incidence. Unfortunately, many cases of actinomycosis, particularly in the cervico-facial area, are treated on an out-patient basis and misleadingly low figures are therefore attained using in-patient records. Hence Cope's figures (1930) gave an incidence of 1.7 (mean annual) using 167 cases from eleven English hospitals over a period of some 97 years in total, whereas Porter arrived at a figure of 7.5 using 98 cases over a period of 13 years in the North East of Scotland. Clearly, these figures must be regarded with some reservation.

Site

Actinomycosis is characteristically found at certain sites in the body - namely Cervico-facial, Respiratory and Abdominal. Israel classified cases up to 1885 according to the point of entry thus:-

(a) Cases of invasion through the Oral and Pharyngeal cavities.

(b) Cases of primary disease of the respiratory tract.

(c) Cases of primary disease of the intestinal tract.

(d) Cases with uncertain point of entrance.

Most studies clearly indicate that cervico-facial actinomycosis is the most common form of the disease (Table 2, 1) with abdominal the next most prevalent and thoracic the least. Cope's series of 1330 cases in 1938 showed 63% to be cervico-facial, 22% abdominal and 15% thoracic. Cruikshank (1969) noted that 65% were cervico-facial and abdominal the next most prevalent (19%) and thorax and skin accounting for the remainder. Amongst the other surveys of the incidence of cervico-facial actinomycosis, Porter (1951) attributed his relatively high figure to the fact that the dental department of Aberdeen Royal Infirmary contributed 31 cases to his figures. Both Lentze (1961) with 93% cervico-facial and Holm (1950) with 90% cervico-facial had close ties with dental departments. On the other hand, Brown's survey in 1973 showed only 32% cervico-facial cases. Nevertheless, it is apparent that the disease is most often in this area of the body, reflecting the endogenous source of the infecting micro-organism in the oro-pharyngeal region. The distribution of lesions within the cervico-facial group is difficult to elucidate clearly. Goldstein et al (1972) went so far as to state that cervico-facial actinomycosis was virtually synonymous with mandibular and cervical involvement. From perusal

of the many case reports published, this contention is clearly well founded. In the mandibular and cervical area, the most prevalent site would appear to be the submandibular region. Baranczak (1971) noted that 148 of 264 cervico-facial cases (60.7%) were in the submandibular area. Other common sites are the body of the mandible (Bramley & Orton 1966) the neck, cheek and submental region. Involvement of the maxilla is considered rare, and probably reflects the better blood supply and drainage of this region which renders it less liable to infection generally. Davis & Voelker (1925) in a review of 670 cases in the United States of which 400 were classed cervico-facial, reported only 2 involving the maxilla (0.5%) while Glahn (1954) reported 1 case from 90 cervico-facial lesions (1%). Tokiwa et al (1959) reported an 8% maxillary involvement in their review of 283 cases in Japan. Goldstein et al (1972) in a general review of actinomycosis of maxilla indicated that it was more often seen as a localised intra-oral infection than as a typical cervico-facial presentation, and this is borne out by cases reported by Main & MacPhee (1964) of a periodontal abscess, and Hammer & Schaefer (1965) and Browne & O'Riordan (1966) of periapical lesions.

Related to the maxilla but considered separately are cases involving the maxillary air sinus. Stanton (1966) reported a case of bilateral actinomycosis of

the maxillary sinuses and reviewed reports of 7 previous cases. Of these 7 cases, 4 were a direct result of dental infection following extractions.

Salivary glands may also be involved, notably the parotid gland. Sanford & Magath (1921) noted 1 case involving the parotid and 1 which affected the parotid and the spinal cord in their 96 cases in the Mayo Clinic. Hopkins (1973) reported a case of primary involvement of the parotid and Sazama (1965) reported 5 cases of parotid actinomycosis of which 4 were extensions of cervico-facial actinomycosis resulting from dental infection. Killey et al., (1971) reported one case of actinomycosis of the submandibular gland in which there was no history of extraction, and which was provisionally diagnosed as a carcinoma.

Separate reports have also been made of involvement of the temporomandibular joint (Bradley 1971, Breuer 1951) but these cases generally arise as a complication of cervico-facial disease. In the case reported by Bradley, however, the infection was primary in the joint.

Age

Actinomycosis may occur at any age and cases have been reported as young as 2 years, (Porter, 1951) and as old as 82 years (O'Mahoney, 1966). However the maximum age incidence would appear to be in the 21-30 age group. Cope (1952) considered that

the majority occurred between 15 and 35 years and indicated that less than 3% occurred under the age of 10 years. Porter (1951) compared the population spread with the incidence, and noted that in the 11-20 and 21-30 age groups, there were more cases than would be expected proportionally. In general, the age incidence of all cases of actinomycosis and cervico-facial cases specifically bears a close relationship.

It is difficult not to equate the age incidence of the disease with the presence of the teeth which harbour the causative micro-organisms. There is no doubt that the maximum incidence of dental infection occurs in much the same age range and that over the age of 30, the occurrence of the edentulous population at least in this country rises sharply.

Sex

Comparison of the various surveys and reports produces a remarkably constant figure of males being affected in a ratio of 2:1 compared with females. (Table 2, 2) Certainly trauma in the form of facial bone fracture is more common in the male, but this would not explain the wide difference in sexual occurrence. If females are more dentally conscious than males, this might influence the oral hygiene and hence the bacterial population. In the Adult Dental Health Survey in Scotland (1972) the oral debris assessment and calculus score showed females to have a consistently lower incidence of both debris and calculus.

Reference		% Age Cervico Facial	Number of Cases
Sanford and Magath	(1921)	66.6	96
New and Figi	(1923)	68.1	107
Davis	(1944)	50	46
Davis and Voelker	(1925)	59.7	670
Porter	(1951)	75	95
Holm	(1950)	90	224
Brown	(1973)	32	181
Lentze	(1961)	93	1002
Cope	(1938)	63	1330

Table 2, 1. Percentage cervico-facial actinomycosis of total numbers of cases.

Reference		% Age males	Number of cases
Sanford & Magath	(1921)	84	96
Porter	(1951)	69	98
Tokiwa et al	(1959)	70	283
Baranczac et al	(1971)	72	264
Foulerton	(1913)	65.4	78

Table 2, 2. Percentage male cases in cervico-facial actinomycosis.

Chapter 3

The Clinical Features of Cervico-facial and Intra-oral Actinomycosis

James Israel in 1878 published a report of two cases of actinomycosis affecting human beings. They both had the symptoms of "chronic pyaemia" and both had histories of trauma to the affected part. The first concerned a 39 year old female who developed the disease after a breast injury and which affected the left thoracic wall, and the second was a 36 year old male with cervico-facial actinomycosis. The latter patient had a history of intra-oral swelling involving the cheek teeth on that side in the previous year. He subsequently developed a submandibular swelling (like a lymph gland) which was initially painless. Over a period of about one month the swelling enlarged, became progressively more painful, and eventually involved the whole of the right side of the neck. The swelling was noted to be red with induration of the surrounding tissues, and it required to be "lanced" on three separate occasions. The discharge was described as "profuse, stinking" and "strewn with yellowish millet seed sized grains."

This very early description of the disease leaves little to be added, and it remains a classic description of the disease in man.

As has been discussed in Chapter 2, cervico-facial actinomycosis may present in a variety of sites around the face and jaw region. Most commonly, however, the swelling is situated in the submandibular region or in

the area related to the body and lower border of the mandible. Other principal sites are the cheek, the parotid/masseteric area, and the submental region. In the classic case, the swelling is brawny, hard, and produces a bluish dusky tinge on the skin surface. If left untreated, it tends to discharge onto the skin through multiple sinuses. The infection has a predilection for connective tissues, but rarely passes along the recognized planes. The infection progresses by random involvement of adjacent areas, and the sinuses often close rapidly after discharging, only to break through skin again in the freshly involved region (Cope 1952). The pus may contain tiny yellow grains known as "sulphur granules" but this is not necessarily a constant feature of the disease, particularly in the early stages. There is an undoubted tendency for the facial or cervical infection to gravitate towards the mediastinum (Aird 1958). The adjacent unaffected tissue walls off the disease by a fibrotic reaction, and this may lead to unsightly scarring on resolution. However, an "atypical" actinomycosis has been recognized by many workers which presents in an acute or sub-acute manner. Axhausen (1935) was an early worker to note this form of the disease, relating it to the tissue reaction to the accompanying pyogenic bacteria. Sullivan and Goldsworthy (1940) and Goldsworthy (1947) suggested that in the acute form of the disease,

suppuration predominated over fibrosis, and that the lesion became indistinguishable from any other acute dental abscess. Glahn (1950, 1954) was probably the first to divide cases definitively into the "classic" and "atypical" forms, on the basis of acuteness. He supported Holm's contention that the infection was a combination of actinomyces with other synergistic bacteria, and that the atypical acute case was a result of the particular combination of organisms involved. More recently Mitchell (1966), and Bramley and Orton (1960) defined an acute or subacute group on broadly similar lines.

Pain is a variable feature of the disease. It was largely this symptom and the location of the swelling which prompted Bramley and Orton (1960) to divide their 11 cases into 6 acute painful cases and 5 chronic slow growing cases in which pain was not a feature, and in which the swelling arose at the lower border of the mandible at the point where the facial vessels cross. Mitchell (1966) also laid emphasis on the co-relationship between the acuteness of the infection and the presence of subjective symptoms. In his post traumatic group (23 of 46), the pain varied in intensity with 13% describing it as severe while no fewer than 87% experienced only mild discomfort or no pain at all.

Hertz (1957, 1960) recognized the classic cervico-facial actinomycosis and the "atypical" form of Glahn. The former he considered characteristically painless whilst the latter he described as acute or subacute and resembled "ordinary dental inflammation." He proposed a further group of atypical cases characterised by being painless, though slightly tender to palpation and being firmly adherent to bone but having no osseous focus. These lesions, he claimed, did not yield Actinomyces israelii but a "species of actinomyces." A common co-existing bacterium was an anaerobic streptococcus.

Human actinomycosis is a disease predominantly of soft tissues. Bone is involved infrequently in marked contrast to the corresponding disease in animals (lumpy jaw). Nevertheless reports have been published of actinomycosis directly affecting bone. Cope (1952) reported massive thickening of the mandible by interstitial and periosteal bone formation. He further noted the osteolytic destruction of bone related to an infected tooth which eventually presents as an external facial or cervical swelling.

Nathan et al in 1962 reported 4 cases, 3 of which affected the bone of the mandible and one the maxilla. They indicated that the radiographic appearance of such cases varied from lytic destruction without reactive bone formation to a marked sclerosis. They stressed the rarity of true bony infection by actinomyces and

that the radiographic changes recorded were not specific to the disease.

Trismus is another common feature of cervico-facial actinomycosis (Cawson 1968; Killey et al 1971; Norman 1970; Hertz 1957). Cope (1952) noted that this symptom may be quickly manifested and may persist for a considerable period even after resolution of the swelling. Mitchell (1966) noted that trismus was a more constant and troublesome feature in the acute or subacute presentation and Bates (1933) considered trismus to be an invariable feature of his 17 reported cases.

Pyrexia is variable and tends to be related to the acuteness of the infection. Mitchell noted that all his acute group had temperatures over 99°F whereas 82.6% of his post-traumatic chronic group were recorded at or below 99°F. Generally, however, pyrexia is not a prominent feature of cervico-facial actinomycosis but may be more marked in the other forms. Brown's survey of 181 cases (1973) (of which only 33% were cervico-facial) indicated that fever was present in 89% of cases and was 100-104°F in 66%.

An interesting feature of the disease is the lack of involvement of lymph nodes. Lymphadenopathy is rarely found (Thoma 1970), and direct involvement of nodes is an extreme rarity. Mitchell (1966) found that lymphadenitis was more commonly associated with the more acute form of the disease. Cawson (1968)

considered that although primary involvement was infrequent, secondary infection may cause lymphadenitis.

Intra-oral Actinomycosis

This form of the disease differs from cervico-facial actinomycosis in that it does not produce the external facial swelling, is often found by chance pathological examination of fixed tissue specimens, and does not generally require protracted antibiotic therapy to effect resolution.

Intra-oral actinomycosis may conveniently be sub-divided into cases directly involving the dentition and those which involve the soft tissues of the oral cavity. The former is the larger group, the latter being composed mainly of actinomycosis of tongue. When discussing intra-oral actinomycosis one enters a "grey area" in which the pathogenic role of the micro-organism must be, in some cases, open to doubt. Nevertheless, the aetiological significance of such cases in respect of the classical cervico-facial actinomycosis can be clearly inferred.

Actinomyces may be traced from carious lesions to dental pulps and root canals and to apical granulomata and cysts. Howell et al in 1962 noted that A. israelii was the predominant organism (isolated in culture) in a high percentage (40-50%) of dental plaques from either shallow or deep carious lesions. It should be noted, however, that both Morris (1954) and Ennever et al (1951) reported that

they could not correlate the occurence of actinomyces in the mouth (mainly dental plaque) with the incidence of dental caries itself.

Actinomyces have been noted in the dental pulp (Hardwick and Newmann 1955; Villa 1957) in clusters of Gram positive rods radially arranged, and have been isolated from infected root canals. Shovelton and Sidaway in 1960 isolated 19 stains of anaerobic actinomyces from 110 teeth in which no previous contamination of the canals had occurred by dental treatment. Several authors have reported actinomycotic colonies associated with apical granulomata (Browne & O'Riordan 1966; Hamner & Shafer 1965; Kalnins 1971; Gee & Sullivan 1940; Goldstein et al 1972; Kapsimalis et al 1968; August & Levy 1973; Gold & Doyne 1952). In all these reports the diagnosis was made by pathological examination of fixed specimens. The symptoms and signs were indistinguishable from the clinical features of these lesions caused by other bacteria. The organisms have also been found in dental cysts (Sprague & Shafer 1963; Martinelli & Rulli 1967).

A large proportion of the cases of maxillary actinomycosis are localised periapical lesions (Hamner & Shafer 1965; Kalnins 1971; Browne & O'Riordan 1966; Goldstein et al 1972; August & Levy 1973) which seldom attain the proportions of the mandibular infections.

Actinomycosis of tongue must be considered uncommon. Burket (1957) cited at 3.7% incidence and Thoma (1970) 3%. Sporadic cases are reported on occasions, (Dorph-Peterson & Pindborg 1954; Cameron 1932) and a comprehensive report of 12 cases was given by Figi in 1926.

Cope (1952) and Thoma (1970) both noted the predilection for the anterior two thirds of tongue and the fact that the lesions tended to be confined to the tongue substance, not spreading to adjacent regions. It had to be differentiated, Cope noted, from malignant neoplasm, tuberculosis and the gumma of tertiary syphilis.

Materials and Method

Cases of cervico-facial and intra-oral acinomycosis diagnosed in Glasgow Dental Hospital over the period 1968-75 were reviewed. Details of clinical presentation and history were obtained in an attempt to classify the cases in the light of previous reports. A total of 15 cervico-facial cases and 11 intra-oral cases were investigated. (See Appendix A. & B.)

Results

Cervico-facial actinomycosis Table (3,1)

The average age of the 15 cases was 26.9 years with a male female ratio of 6 to 9. Five cases were related to dental extraction and in this group, the average age was 35.6 years. All the teeth involved

were lower molars and the interval between extraction and incision ranged from 6½ weeks to 7 months. Only two cases (Cases 12 and 13) had no direct dental link.

Seven cases were associated with dental abscesses of which 6 were lower molar teeth and 1 was a premolar root. (fig. 3, 1 and 3, 2) The average age of these cases was 19.3 years. This is younger than the post-extraction cases, and comparison of the age incidence of these two types of presentation by Mann-Whitney U test indicated that this age difference was highly significant ($P<0.01$).

Of the 15 cases, 10 were described as acute swellings and 5 as either sub-acute or chronic. All the cases related to a dental abscess were of the acute type of presentation while those following trauma were all described as sub-acute or chronic in character. (Figs. 3, 2 to 3, 9)

Pain was noted as a common symptom of the acute type of actinomycosis, being a noted feature in 9 of the 10 cases. (Table 3, 2) It was also a feature of the cases involving the parotid gland and the right cheek. Trismus was recorded in 6 cases, (Table 3, 2) but showed no predilection for acuteness being more closely related to the site and extent of the infection.

Intra-oral cases (Table 3, 3). No common clinical feature was noted in this group, and no attempt was made to group or classify them with regard to age or sex.

Table 3, 1. The age, sex and site distribution of cervico-facial cases together with related dental focus or association with trauma.

Case	Age	Sex	Site	Associated Focus	Post Trauma	Time Lapse
1	11	F	Left Submandibular	⌈6 abscess		
2	13	F	Left Submandibular	⌈6 abscess		
3	17	M	Right Submandibular	7⌋ abscess		
4	27	M	Left Submandibular Left Submental		⌈7	7 weeks
5	26	F	Right Submandibular	7⌋ abscess		
6	20	F	Right Submandibular	7⌋ abscess		
7	44	F	Right Submandibular		8⌋	9 weeks
8	46	M	Left Submandibular		⌈78	6.5 weeks
9	29	F	Right Body		8⌋	12 weeks
10	40	F	Left Submandibular	⌈5 abscess		
11	34	M	Right Submasseteric	8⌋ Pericoronitis		
12	22	F	Right Parotid			
13	34	M	Right Cheek		? denture	12 weeks
14	8	F	Left Body	⌈6 abscess		
15	32	M	Submental		(a) ⌈56 (b) Curettage left mandible	7 months 6 months

Case	Site	Acute	Subacute/Chronic	Pain	Trismus
1	Left Submandibular	+		-	+
2	Left Submandibular	+		+	-
3	Right Submandibular	+		+	-
4	Left Submandibular Left Submental		+	-	-
5	Right Submandibular	+		+	+
6	Right Submandibular	+		+	-
7	Right Submandibular		+	-	-
8	Left Submandibular		+	-	+
9	Right Body		+	-	-
10	Left Submandibular	+		+	-
11	Right Submasseteric	+		+	+
12	Right Parotid	+		+	+
13	Right Cheek	+		+	+
14	Left Body	+		+	-
15	Submental		+	-	-

Table 3, 2. Relationship of pain and trismus to the acuteness of the infection.

Fig. 3, 1. Peri-apical radiograph of Case 2 showing radiolucency associated with both roots of /6.

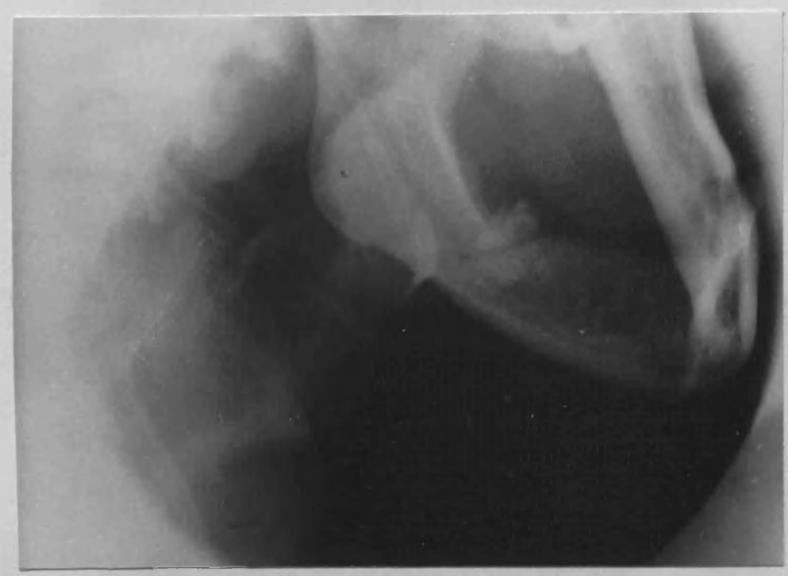

Fig. 3, 2. Lateral oblique jaw radiograph of Case 6 showing $\overline{7}$/ root and apical radiolucency.

Fig. 3, 3. Typical clinical appearance of the acute form of cervico-facial actinomycosis (Case 1).

Fig. 3, 4. Typical clinical appearance of the acute form of cervico-facial actinomycosis (Case 14).

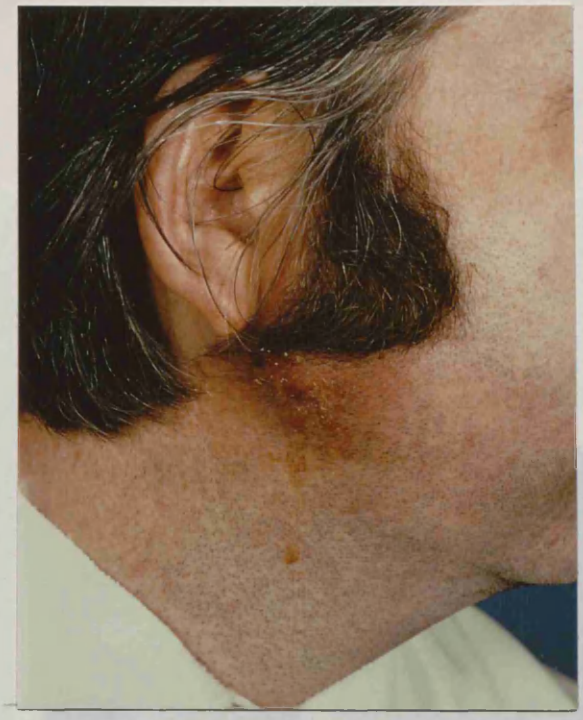

Fig. 3, 5. Actinomycosis presenting as a submasseteric abscess. The organism gained entry to this region from a pericoronal infection of $\overline{8}/$ (Case 11).

Fig. 3, 6. Actinomycosis presenting as a submasseteric abscess. The organism gained entry to this region from a pericoronal infection of $\overline{8}/$ (Case 11).

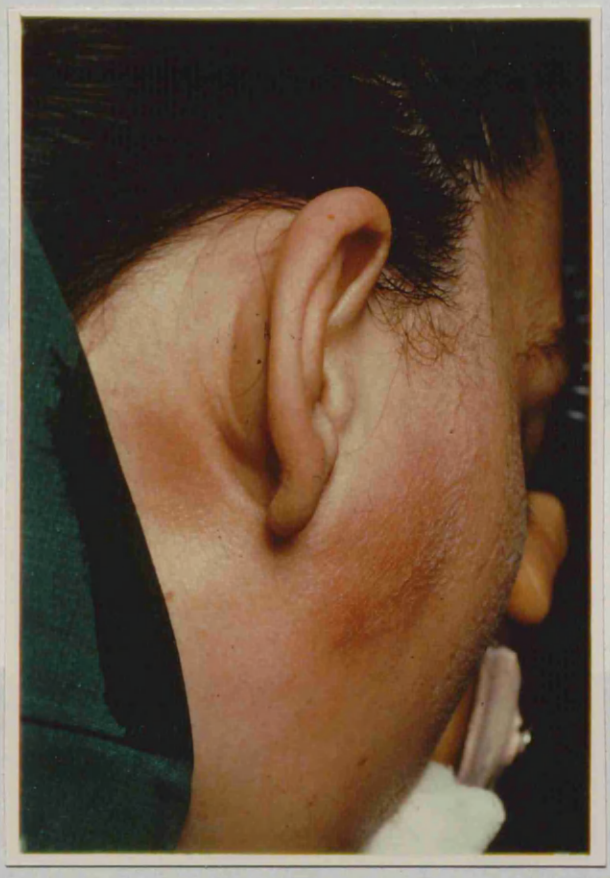

Fig. 3, 7. Acute swelling of parotid gland and masseteric region due to actinomycosis (Case 12).

Fig. 3, 8. Chronic type of actinomycosis presenting as a submental abscess with multiple sinus formation (Case 15).

Fig. 3, 9. Chronic type of actinomycosis presenting as a submental abscess with multiple sinus formation (Case 15).

Case	Age	Sex	Associated Lesion	Diagnosis
A	39	F	Infected Odontogenic Keratocyst $\overline{2 \cdot\cdot \vdash \cdot\cdot 2}$	Path
B	40	F	Periodontal Abscess $\lfloor\underline{45}$	Path
C	6	F	Pulpitis $\underline{5}\rfloor$	Path
D	21	F	Infected Apical Dental Cyst $\underline{6}\rfloor$	Path Bact
E	65	F	Ossifying Fibroma	Path
F	76	M	Paget's Disease	Path
G	18	F	Compound Composite Odontome $\overline{7 - 4\rfloor}$	Path Bact
H	41	M	Dental Cyst $\overline{21 \mid 12}$	Path
J	25	F	Chronic Abscess Left Cheek	Path
K	70	F	Residual Dental Cyst $\_\rfloor$	Path
L	35	F	Lympho-Epithelial Cyst Right Floor of Mouth	Path

Table 3, 3. Intra-oral cases indicating associated pathology and method of diagnosis.

Discussion

Subdivision of cervico-facial actinomycosis has been proposed by several authorities using varying criteria. Most authors (Mitchell 1966; Bramley & Orton 1960 Hertz 1957, 1960; Glahn 1950, 1955) have differentiated acute actinomycosis as a separate clinical entity. This contention is well supported by the present study of which 10 were so described. The reason for this variation in presentation most probably lies in the mixture of bacteria which are found in actinomycosis, their proportions, virulence and the host reaction to them. Mitchell (1966) went as far as stating that the acute form associated with an infected tooth was not true actinomycosis but a simple pyogenic dental abscess in which actinomyces were present. There is, however, a general acceptance that infections in which actinomyces are isolated are actinomycotic and should be so regarded. Hertz (1957, 1960) described another atypical actinomycosis which he stated was a painless swelling firmly adherent to the mandible. Two cases in this review (Cases 7 and 9) could be so described but no significance can be placed on such a clinical variant.

There can be no doubt of the importance of the teeth in the infection. The cases which followed dental extraction were found to be chronic in type, less painful and affected a significantly older age group. Those cases with a related infected tooth were acute in

character, generally caused more pain, and were seen in a younger age group. In both these main groups, however, the predominant area was the lower molar region. This site predominance agrees closely with most reviews of cervico-facial actinomycosis. Mitchell (1966) noted that in 42 of 43 cases with dental association, the lower posterior quadrants were implicated. Rud (1967) in a review of 10 cases noted that 8 involved the lower third molar region and O'Mahoney (1966) and Bramley & Orton (1960) also support this finding.

The average age of the post trauma group (35.6 yrs.) corresponds with other reports such as Rud (1967) with an average age of 34.5 from 4 cases, Holmes (1958) 42.4 yrs. from 5 cases and O'Mahoney (1966) 42.3 yrs. from 6 cases.

In terms of the swelling, no specific difference was noted between actinomycosis and other dental infections. In only two cases (Cases 8 and 15) did the patients present with the multiple draining sinuses and granules evident in the discharge. In all other cases, the swelling was surgically opened, and this reflects earlier presentation of these patients for medical help.

Involvement of bone in cervico-facial actinomycosis is rare (see introduction) but one case (Case 8) demonstrated indirect involvement of the angle of the mandible by erosion. (Fig. 3, 10) This case which

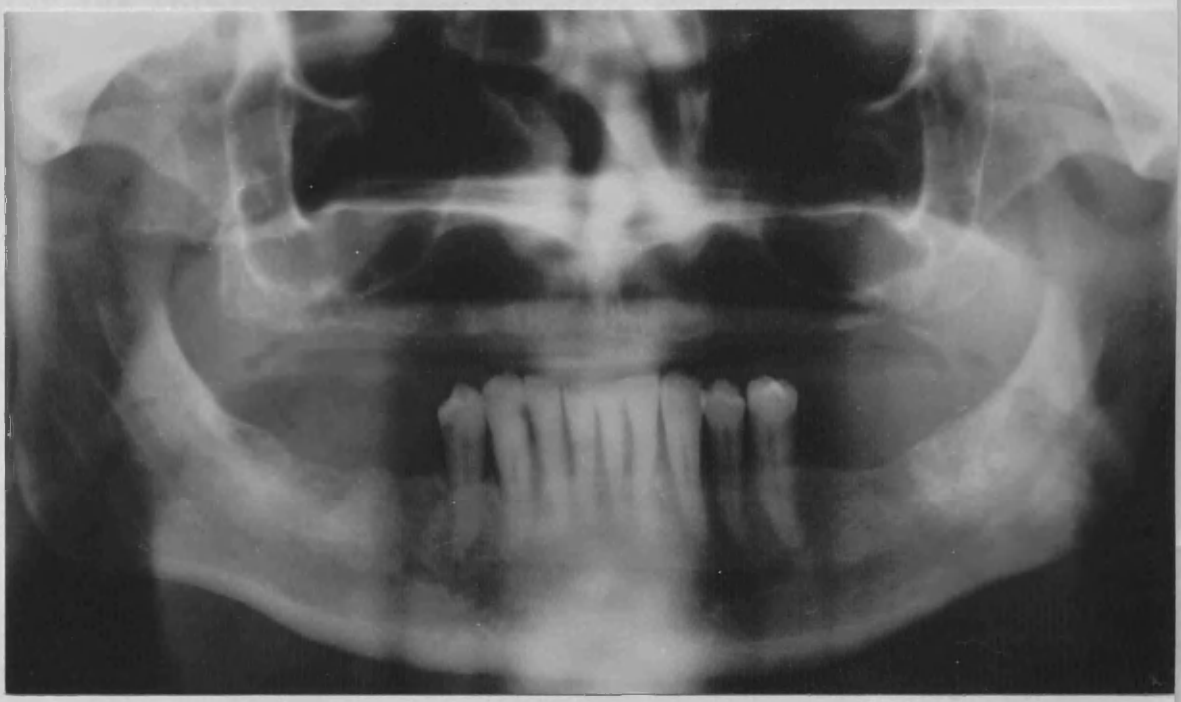

Fig. 3, 10. Orthopantomograph of Case 8 showing erosion of the left angle of mandible (compare with Fig. 5, 2).

which recurred after initial treatment highlights other features of the disease process. Firstly, the reluctance of the patient to seek attention indicates the lack of severe subjective symptoms in the non-acute form, and secondly the tendency for the infection to persist after therapy. (See Chapter 5)

The tendency of the infection to move or gravitate from its original site has been noted (Aird 1958). Two cases in the study (Cases 4 and 15) displayed this characteristic. In both, the infection moved from the submandibular region as chronic painless swellings until eventually presenting in the submental area where the pus tracked to the surface. The original submandibular site resolved totally and required no surgical intervention. Had the infection been more acute in type, it is doubtful whether this "migration" would have been possible, and the phenomenon would thus appear to be a feature of the chronic actinomycosis.

Cases of intra-oral actinomycosis were, with few exceptions, chance findings. They did not lend themselves to grouping and were totally indistinguishable from non-specific oral infections clinically. It is of interest, however, to note that in 7 of the cases, there was some physical obstruction to the micro-organism gaining access to the soft tissues. It would appear that where the infection is limited by an epithelial or

fibrous tissue lining, or some calcific barrier, such as a pulp chamber or osteosclerosis around a periapical esion, there is less likelihood of the classic cervico-facial form of the disease developing.

Conclusions

Cervico-facial actinomycosis may be classified as dental or non-dental in origin, the vast majority being directly related to the dentition, particularly the lower posterior quadrants. Cases may also be classified on the basis of relationship to an infected tooth or subsequent to trauma in the region, usually extraction. Finally, the acute actinomycosis would appear to be a distinct clinical entity, and this study indicates that those cases related to an infected tooth were predominantly acute in character.

The majority of cases did not present in the textbook manner, and the implication of this non-specific mode of presentation to diagnosis of all facial swellings is apparent.

The intra-oral cases highlight the ubiquitous nature of the bacteria, and serves to underline the need for careful laboratory examination of all oral pus specimens.

Chapter 4

The Diagnosis of Actinomycosis

Introduction and Review of Literature

The diagnosis of cervico-facial and oral actinomycosis depends upon two disciplines. Firstly, the clinical diagnosis of the infection which depends upon the clinician's high suspicion index when confronted by any infective swelling in the region, and the second is the laboratory diagnosis which implies the definitive confirmation of the clinician's suspicion.

Recognition of actinomycosis by dental surgeons and physicians alike is probably poor. Brown's study of 181 cases (1973) revealed that in only 19 cases did the clinician correctly diagnose the condition, although the lesions were commonly noted to be inflammatory masses or abscesses. This is not totally surprising, since the disease is not common (Chap. 2), but it does highlight the need for constant vigilance and an awareness of the possibility of actinomycosis. Glahn (1954) considered that all abscesses in the soft tissues related to the jaws or teeth should be suspected as being actinomycotic until proven otherwise. Sims (1974) in a study of 1,000 consecutive pus specimens from dental sources found actinomyces in 57 cases of which only 8 were "full blown" cases. He was of the opinion that any purulent oral lesion should be regarded as an early case of actinomycosis, and maintained that if these 57 lesions had been allowed to persist, then the actinomyces would have proliferated to a more recognizable disease. Most authorities stress the importance of the chronicity and

granulomatous tissue reaction of the lesion. Kruger (1968) considered the presence of multiple draining sinuses to be important, although this in itself implies late diagnosis of the disease. Goldstein et al (1972) in a discussion of intra-oral actinomycosis proposed that any chronic localised periapical infection associated with trauma or dental manipulation, such as endodontic therapy, was suggestive of actinomycosis. The examination and collection of any pus discharge is universally emphasised. Lesney & Traeger (1959) advocated the use of the hand lens and the dilution of pus to detect sulphur granules. The presence of granules however is variable (Sims 1974) though their presence must be strongly suggestive of the disease (Kruger, 1968; Cruikshank, 1969; Rud, 1967). The handling and disposition of the pus is important. Hylton et al (1970) considered that the use of routine swabs or smears rendered the characteristic filamentous morphology of the actinomyces unrecognisable. They urged the use of a curette to transfer specimen material from the wound to a glass slide. Rud (1967) insisted that the pus should be aspirated or taken by aseptic incision. There is no doubt, however, that as with all pus samples, as much material should be collected as possible, preferably in a sterile Petri dish or test tube, and that it should be sent to the bacteriology laboratory as quickly as possible. Many cases of actinomycosis are diagnosed by pathologist's scrutiny of fixed tissue specimens.

Glahn (1955) pointed out the disadvantage of such diagnoses, and cited that only 1 of 17 proven (bacteriologically) cases was successfully biopsied.

Laboratory Microbiological examination of pus is the essence of accurate diagnosis of actinomycosis. Goldsworthy (1947) stated that a diagnosis could not be made with certainty by any other means. The examination of the pus involves two stages. Firstly, the direct examination of any granule which has been found by Gram stain, and the second involves culturing the pus both aerobically and anaerobically. Cruikshank (1969) considered the finding of Gram positive branching mycelia sufficient for clinical diagnosis and many case reports confirm that this method of microbiological confirmation is widely recognised as sufficient. (Lesney & Taeger 1959; Hylton et al 1970). However many consider that the culture of the actinomyces is of great importance, and that absolute diagnosis depends upon successful culture. (Thoma, 1970). There is often difficulty in culturing actinomyces from mixed bacterial infections. No selective medium exists, and the best results are usually obtained by thorough washing of the granules and incubating anaerobically both in fluid medium (Brewers Thioglycollate) and on blood agar. Further tests are then possible both to confirm that the bacteria are in fact actinomyces and to allow discrimination of the strain. These involve biochemical reactions, fluorescent tests or cell wall analysis.

(Videinfra) Fluorescent antibodies have been used directly on smear preparations and to allow rapid monitoring of mixed cultures of bacteria, (Slack et al 1960; Blank & George 1968). These techniques have the advantage of speed, and hence early accurate diagnosis.

The value of the biopsy has been noted as rather unreliable. Examination of fixed tissue specimens in paraffin sections has obvious limitations. Multiple sections may be required to include a sulphur granule, and if the clinician has no suspicion of actinomycosis, the pathologist might not "ribbon" his material in a search for the granules. Furthermore, particularly in lung or subcutaneous tissues, nocardiosis is extremely difficult to differentiate from actinomycosis. The fundamental difference between the organisms is that nocardia are aerobic whilst actinomyces are essentially anaerobic or micro-aerophilic. Some clarification is claimed by Hotchi & Schwarz (1972) who used fluorescent antisera on paraffin sections of tonsils with actinomycotic granules with some degree of success.

Diagnosis of Actinomycosis

Materials and Method All cases of actinomycosis reported from 1968 in the routine microbiological and pathological services in Glasgow Dental Hospital were reviewed to ascertain the method of diagnosis in each case. Both cervico-facial and intra-oral cases were included in the study. Fluorescent antibody stain

testing (Lambert et al 1967) was implemented in the diagnostic microbiological department in an attempt to evaluate the use of such material for routine diagnostic purposes.

The fluorescent antibody serum was obtained from the Biological Reagents Section, National Center for Disease Control, Atlanta. The conjugates used were : A. israelii (serotypes 1 and 2) and A. naeslundii (absorbed with A. israelii cells). In those cases suspected of being actinomycosis clinically, the pus was smeared directly onto a glass slide and a Gram film prepared. If this film showed Gram positive branching filaments or bacilli, a direct fluorescent antibody test was carried out using both conjugates.

The smear was gently heat fixed and the fluorescent serum layered onto the preparation in a moist chamber at room temperature. Incubation was carried out for 30 minutes after which the preparation was washed thoroughly for at least 30 minutes with fluorescent antibody buffer, and mounted in buffered glycerol (9 parts buffer to 1 part glycerol).

Sections were examined with a Wild M 20 research microscope fitted with a dual illuminator and with illumination supplied by an HBO.200W. mercury lamp in conjunction with a F.I.T.C. barrier filter. (Polaron, London) Magnifications of x 10, x 20 and x 40 were employed.

In addition to this immediate examination of

suspected pus, the fluorescent examination was repeated after 3-5 days anaerobic incubation of the pus on blood agar. (95% H. 5% CO_2) Suspected colonies were removed from the anaerobic culture, smeared onto glass slides and stained with the Gram and fluorescent antibody methods.

In addition, a retrospective review was carried out on the results of culturing pus from cases of cervico-facial actinomycosis, in an attempt to ascertain the prevalence of mixed infection and the bacteria which exist in such infections.

Results

Cervico-facial Cases (Table 4, 1) Of the 15 cases of cervico-facial actinomycosis, only one case was diagnosed solely from pathological examination of formalin fixed material (Fig. 4, 1 and 4, 2). Of the 14 cases diagnosed by microbiological means, all showed positive direct films of Gram positive branching filaments or bacilli (Fig. 4, 3), and in only one of these cases were granules not noticed specifically in the pus specimen (Fig. 4, 4). Similarly, in these 14 cases culturing was successful, 11 cases producing the typical colonial pattern of actinomyces israelii. (Fig. 4, 5 and 4, 6) In two cases there was doubt as to the species of actinomyces obtained and in one case, sugar fermentation tests revealed the micro-organism to be actinomyces odontolyticus. (Tab. 4, 2)

Six cases were tested with the fluorescent antisera all of which were positive with direct films

Case	Granules	Direct Gram	Culture	Sugar Fermentation	Direct Fluorescence	Culture Fluorescence	Pathology
1	+	+	+	+			
2	+	+	+				+
3	-	+	+				
4	+	+	+				
5	-	-	-				+
6	+	+	+	+			
7	+	+	+	+			
8	+	+	+	+			
9	+	+	+				
10	+	+	+			+	
11	+	+	+			+	
12	+	+	+			+	
13	+	+	+		+	+	
14	+	+	+		+	+	
15	+	+	+		+	+	

Table 4, 1. Method of diagnosis of cervico-facial actinomycosis.

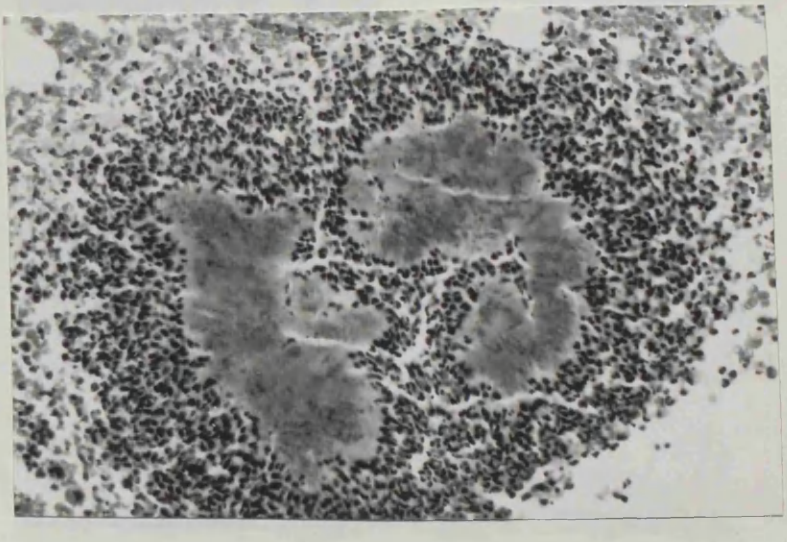

Fig. 4, 1. Photomicrograph of actinomycotic granule from formalin fixed paraffin section (Case 5).
Haematoxylin and Eosin x 384.

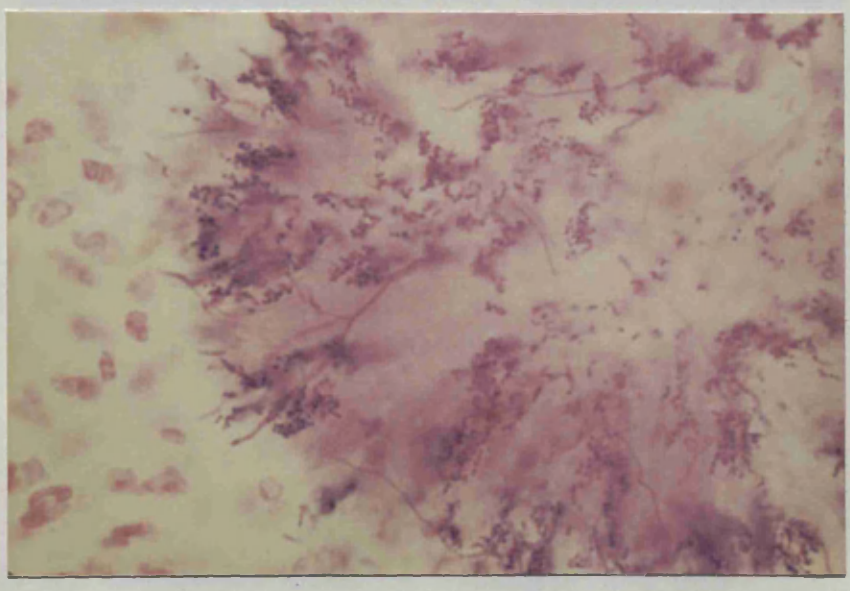

Fig. 4, 2. Photomicrograph of the periphery of actinomycotic granule from formalin fixed paraffin section (Case 2).
Gram stain x 960.

Fig. 4, 3. The periphery of a crushed sulphur granule showing gram positive branching filaments and pus cells.
Gram stain x 960.

Fig. 4, 4. Petri dish of bloodstained pus showing multiple yellow flecks of typical actinomycotic granules (Case 10).

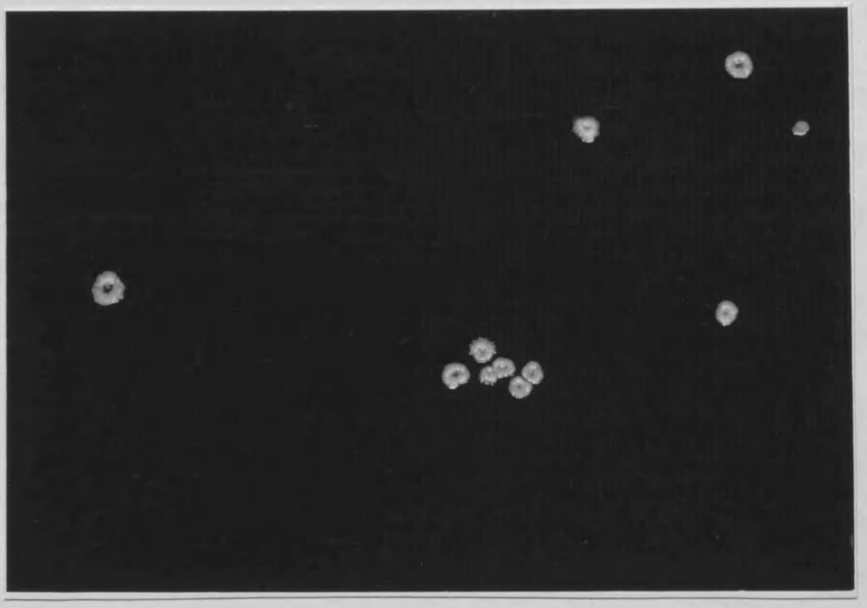

Fig. 4, 5. Low power view of the typical colonial pattern of actinomyces israelii (Case 14).

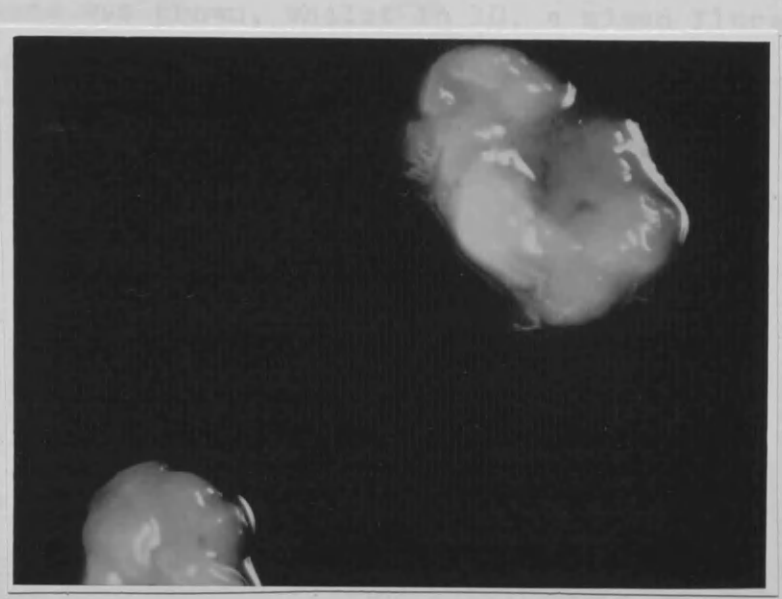

Fig. 4, 6. High magnification of actinomyces israelii colony showing the molar tooth morphology.

of the pus. However, in 3 cases, the value was limited due to the extent of background and non-specific fluorescence. In the other 3 cases, direct smears of the pus were considered positive using the actinomyces naeslundii antiserum as control. (Fig. 4, 7) The films made of the cultured organisms, (3-5 days) were all positive with A. israelii conjugate and totally negative with the A. naeslundii antiserum. (Fig. 4, 8)

Cultures were obtained from 14 of the 15 cases (Tab. 4, 2). In 11 cultures A. israelii was identified positively, in two cases the bacteriologist was unwilling to indicate the species of actinomyces, and in 1 case, A. odontolyticus was identified on the basis of its sugar fermentation reactions. In 3 cultures, pure actinomyces was grown, whilst in 10, a mixed flora was obtained. In 7 of these mixed infections, anaerobic streptococcus was a concomitant micro-organism and this accounts for 53.8% of the 13 cases in which information was available. Eikenella corrodens was isolated in 4 cases (30.7%) and streptococcus viridans and a veillonella species each in 3 cases (23%).

Comparison of the concomitant bacteria with the acuteness of the infection (Tab. 3, 2) revealed no relationship, with anaerobic streptococcus found in 2 of the 5 chronic cases and 5 of the 10 acute cases.

Intra-oral Cases (See Table 3, 3)

All the cases were diagnosed by examination of paraffin sections from formalin fixed material. In

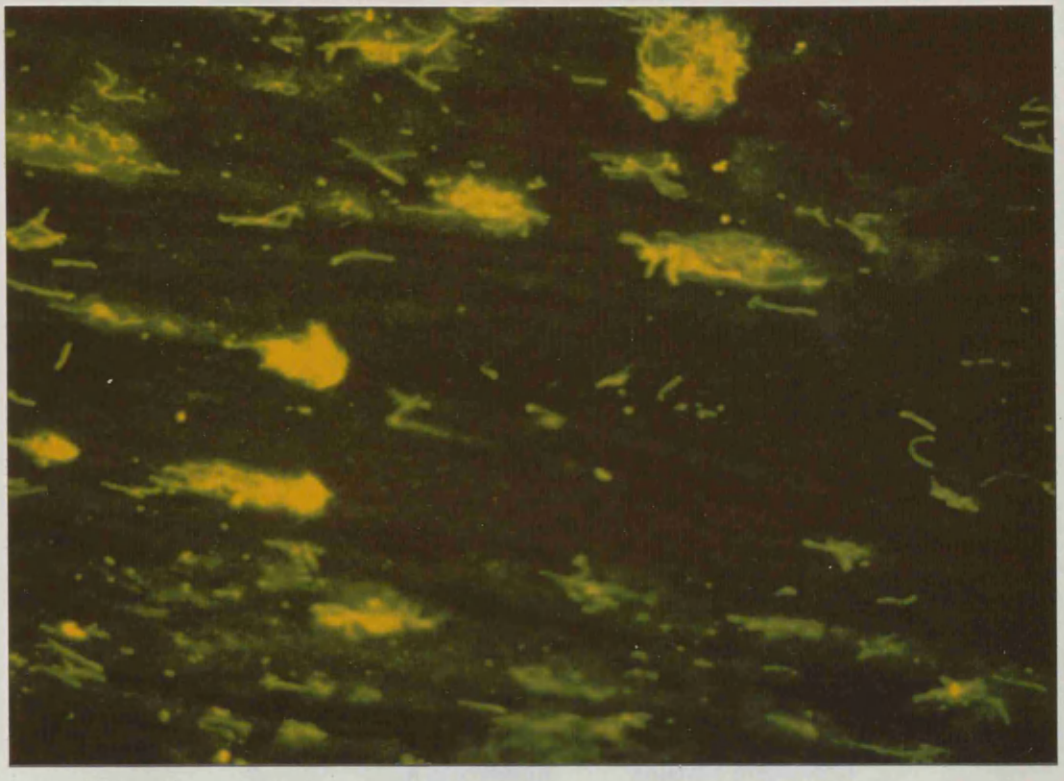

Fig. 4, 7. Direct smear of pus stained with actinomyces israelii fluorescent antiserum x 650 (Case 13).

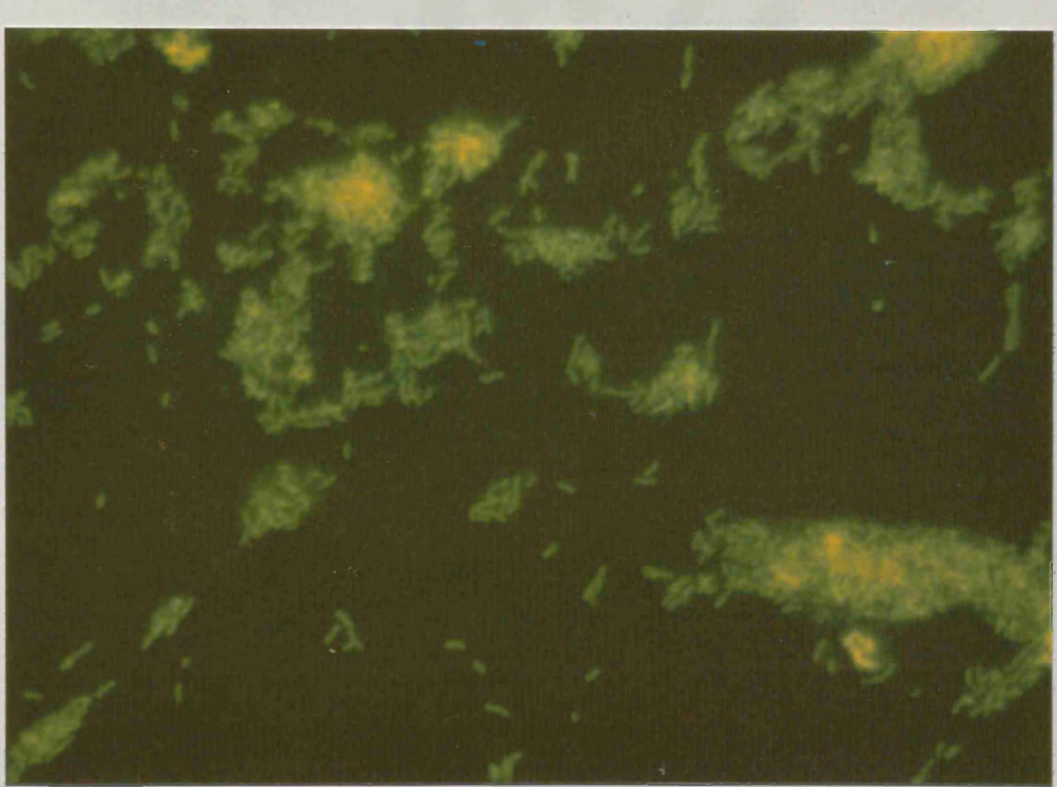

Fig. 4, 8. Smear of culture stained with actinomyces israelii fluorescent antiserum x 650 (Case 13).

Microbiological Cultures from Cases of Cervico-Facial Actinomycosis

Case	Actinomyces	Other Micro-Organisms
1	A israelii	Streptococcus viridans
2	A israelii	Streptococcus viridans Anaerobic streptococcus Unidentified Gram negative bacillus
3	Actinomyces (Species not identified)	Nil
4	Actinomyces (Species not identified)	Staphylococcus aureus Veillonella species
5	No culture	Identified by histopathological examination
6	A israelii	Anaerobic streptococcus Non-haemolytic streptococcus Veillonella species Unidentified anaerobic Gram negative filament
7	A odontolyticus	Anaerobic streptococcus Anaerobic diphtheroid
8	A israelii	Anaerobic streptococcus Streptococcus viridans
9	A israelii	No information
10	A israelii	Eikenella corrodens Anaerobic streptococcus
11	A israelii	Nil
12	A israelii	Eikenella corrodens Veillonella species Anaerobic streptococcus
13	A israelii	Eikenella corrodens Anaerobic streptococcus Bacteroides melaninogenicus Fusobacteria Non-haemolytic streptococcus
14	A israelii	Nil
15	A israelii	Eikenella corrodens

Table 4, 2.

2 cases (Cases D and G, Appendix B) microbiological evidence confirmed the pathological diagnosis, although cultures were not successful in either case. In cases J and L, the bacteria appeared in the typical sulphur granule form (Fig. 4, 9 and 4, 10), but in the other cases the micro-organisms appeared in a less typical form, and were identified by individual morphology and staining. (Fig. 4, 11)

Discussion

The method of diagnosis of 15 cervico-facial and 11 intra-oral cases of actinomycosis in the present study are reviewed. The diagnosis of cervico-facial actinomycosis predominantly by bacteriological investigations stresses the presence of pus in these cases. Intra-oral actinomycosis on the other hand is more often found by chance pathological examination of routine formalin fixed material. In these cases, the pathogenicity of the bacteria and indeed their identification cannot be regarded as reliable. In certain instances, microbiological tests might have clarified such doubt.

The use of fluorescent antiserum was found to be valuable. As was noted, direct Gram films of the pus allows a preliminary diagnosis to be made within minutes of the collection of the specimen. However, delay in confirmation occurs if the organisms require to be cultured, then tested biochemically. This delay may be up to 6 weeks during which time the patient

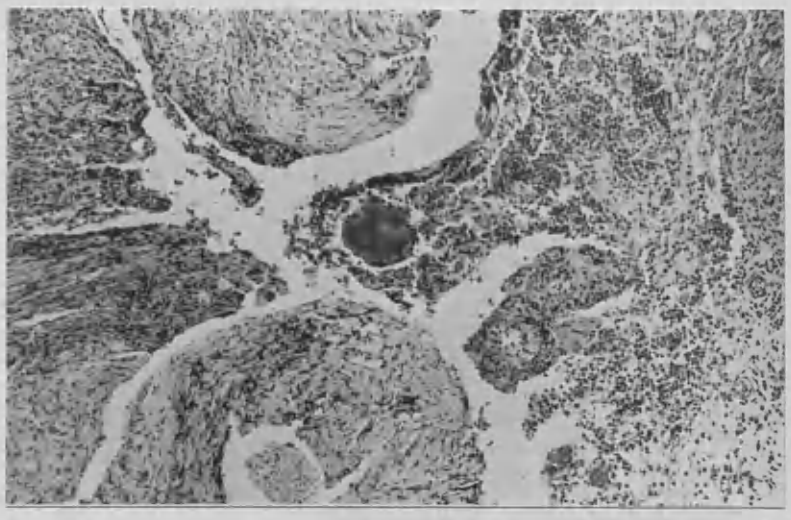

Fig. 4, 9. Sulphur granule surrounded by an inflammatory cell exudate. Haematoxylin and Eosin x 96 (Case J).

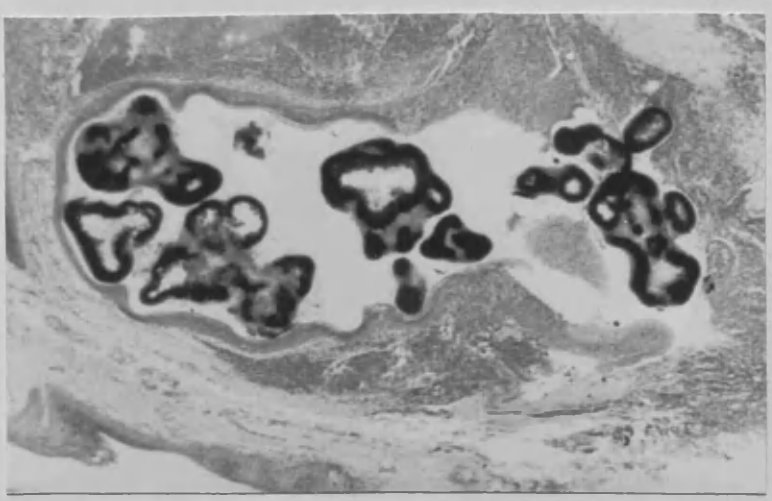

Fig. 4, 10. Sulphur granules lying within a lympho-epithelial cyst. Periodic acid schiff x 24 (Case L).

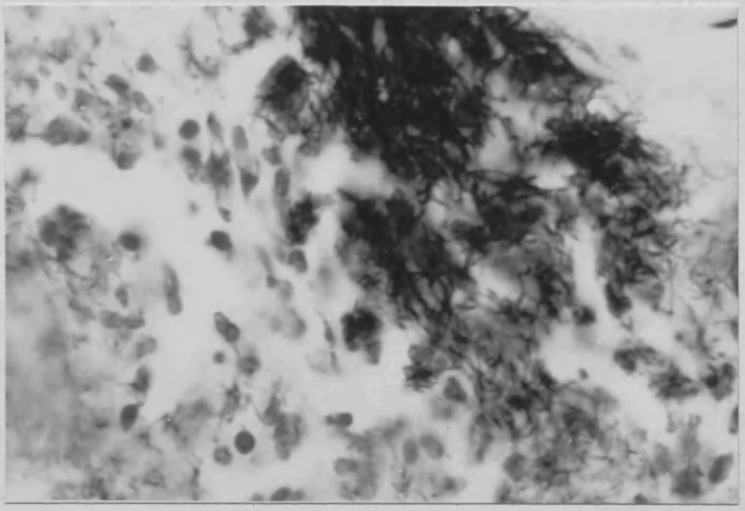

Fig. 4, 11. Histologic section showing branching mycelia lying within a keratocyst Haematoxylin and Eosin x 384. (Case A).

should be receiving appropriate therapy. The fluorescent method allows positive identification within at least 3-5 days of the collection of the sample, and in some cases on the same day. A definitive treatment plan is hence possible with minimal delay.

The investigation of the nature of the cultures obtained showed most cases to be due to a mixture of bacteria. The prevalence of anaerobic streptococcus in these infections has been reported previously (Hertz, 1960). Three cases were due to actinomyces with no accompanying micro-organisms which is in conflict with the views of Holm (1950, 1951) who considered that all cases of actinomycosis were mixed infections. However, in these 3 cases, antibiotics had been administered prior to the establishment of drainage, and this may have affected the culturing of the obtained pus. (Appendix A. Cases 3, 11 and 14) The isolation of Eikenella corrodens in 4 cases agrees with the report of Rud (1967) who isolated Bacteroides corrodens in 6 of 10 cases studied. Although Bacteroides corrodens and Eikenella corrodens are distinct species, their differentiation is relatively recent, and this may well indicate a propensity of Eikenella corrodens to actinomycotic infection.

Conclusions

1. Clinicians should maintain a high degree of suspicion of all inflammatory lesions in the

mouth of jaw region with regard to possible actinomycotic infection.

2. All pus samples should be closely examined for the presence of sulphur granules.
3. Infected soft tissue from intra-oral as well as extra-oral sources should be examined by both microbiological and pathological means.
4. Careful washing of suspected granules and immediate crushing and examination by Gram staining is essential to the rapid provisional diagnosis of actinomycosis.
5. Confirmation of actinomycosis is most rapidly and simply achieved by direct fluorescent testing of suspected pus smears and early culture material. This is now routine practice in the Glasgow Dental Hospital and School following these investigations.
6. Cervico-facial actinomycosis is usually a mixed infection but may fail to yield concomitant bacteria on culture.

Chapter 5

The Treatment of Cervico-facial [illegible]

Introduction and Review of Literature

Actinomycosis has been treated by several methods since its first recognition as a disease entity. The primary means of treatment is a combination of surgery and antibiotic therapy, but other forms of treatment such as radiation, vaccine therapy and iodine therapy have also played a role, mainly in the historical context. Certain European countries such as France and Germany still use vaccine therapy extensively whereas in Britain and the United States, this form of treatment is seldom utilised.

Surgery

Surgical treatment of actinomycotic infection is universally advocated. Whilst acknowledging that antibiotics were the mainstay of treatment, Rowe and Killey (1970) emphasised the importance of appropriate surgical intervention. Goldsworthy (1947) considered the main purpose of surgery was the establishment of free drainage of the pus by accurate anticipation. Not only is surgery necessary in the establishment of unimpeded drainage, it is often the means by which the diagnosis is reached.

Surgical treatment, as indicated, normally involves incision of skin to allow drainage, but just as important is the extraction of any associated dental focus of infection. The teeth normally involved in the infection are lower molars and in particular the lower third molar. Mitchell (1966) considered that in the acute group of

presentations, treatment should probably be confined to removal of the cause (i.e. the tooth) provision of drainage where necessary, and a short course of antibiotic if at all. His experience indicated that the rate of resolution was not affected by provision of antibiotic and that surgical treatment alone was hence adequate.

In the intra-oral situation, most cases are noted to heal after surgery alone. This is probably due to the localised nature of the infection - such as a granuloma - which is totally removed by thorough surgical curettage.

Iodides

Just as some form of surgery, however limited, is universally advocated, so is the systemic administration of iodides almost universally condemned. It was at one time considered to be specific for actinomycosis, but this probably arose from the confusion between actinomycosis in man and animals and actinobacillosis in animals for which iodine has specific beneficial effects (Goldsworthy, 1947). Iodine, in the form of its potassium salt, was administered 0.3 - 0.6 ml. thrice daily, often in milk, the alternative being tincture of iodine. Cope (1952) whilst acknowledging its lack of specificity, considered iodide a useful adjuvant to treatment, in as much as it had a value in promoting "absorption of inflammatory products." Iodine is still advocated as a useful therapy in France especially. (Lebourg et al 1967, Freidel et al 1962)

Radiation

Radiation has been used with apparent success by several clinicians. The main effect of the treatment is to soften the board-like swelling which may occur in the chronic case. Sazama (1965) employed x-rays in three of his 5 reported cases of parotid involvement. The dosage varied between 320 rads and 520 rads in total and in a personal communication (in Bronner & Bronner, 1971) reported 2 further cases in which a total of 2,600 and 2,000 rads was administered in a period of 4 weeks and 3 weeks respectively.

Whilst the beneficial effects of this form of treatment may be an advantage, the damage to underlying and adjacent bone must be weighed carefully.

Most authorities now consider the use of radiation therapy as undesirable in the cervico-facial area, particularly as the rate of cure with other measures, mainly antibiotics, is so high.

Vaccine Therapy

This treatment is rarely used in Britain or the United States but still has devotees in Europe and Germany in particular. The rationale of vaccine therapy is that it promotes antibody response by the patient and brings about a more rapid resolution. The most widely used vaccine is the hetero-vaccine of Lentze (1950) who used several strains of actinomyces in 0.5% carbol which was injected subcutaneously at various dilutions into the affected soft tissues. Buchs (1963) advised

intervals of 5 days between injections, with the strength of each injection progressing from 1:1,000 to 1:1. Bronner and Bronner (1971) considered skin testing of the upper arm with 0.1 cc aliquots of dilutions from full concentration to 1:1,000 to be an accurate indication of the starting vaccine strength. The dilution which produced the most marked allergic cutaneous reaction was regarded as being optimum as a base dilution in any given case. However, systemic upsets may result from the vaccine therapy with fever and nausea, and in many cases it is used with an antibiotic which makes accurate evaluation of its effect difficult.

Antibiotic and Chemotherapeutic Substances

The introduction of sulphonamides in the late 1930's and penicillin in the early 1940's revolutionised the prognosis of patients suffering from actinomycosis. At present, sulphonamides are rarely used due to a combination of microbic resistance to the drug, and the excellent results obtained with antibiotics.

Penicillin is undoubtedly the most commonly prescribed drug in the treatment of all forms of the disease, and this drug has received a justified emphasis in the literature. Many other antibiotics have been employed with good results, and have been proposed as more successful or more logical in the treatment of the infection, but the overwhelming majority of cases respond favourably to penicillin which remains the least toxic and well

tolerated antibiotic.

The dose and duration of penicillin therapy remains a contentious issue. The dose recommended by various reports varies from 1-1.25 million units daily (Mitchell 1966, Tyldesley 1969) to 10-20 million units daily (Hartley & Schatten 1973). Most authorities, however, adopt a flexible attitude in terms of the dose. Norman (1970) divided his cases into 3 groups of severity, and accorded appropriate dose for each category, but he emphasised that larger doses should be prescribed without hesitation should the occasion so demand. Hylton et al (1970) recommended 30 million units of aqueous penicillin intra-venously daily for the first three days followed by 3.2 million units orally for 3 months, and Hertz (1960), in advocating 1.5-5 million units daily, stressed the dosage as being of "paramount importance" to bring about a cure. He considered that inadequate dosage made it impossible to control the accompanying microbes, and cited 4 of his 11 cases as having had previous inadequate penicillin treatment. Rud (1967) also noted that 6 of his 10 cases had been treated inadequately with penicillin and had hence recurred.

Duration of therapy as with dosage of penicillin is open to arguement. Rud (1967) employed short courses of 2-3 weeks (often with sulphonamide) while Holmes (1958) recommended continuation of treatment for at least 3-6 months. Most authorities cite a period of 6-8 weeks with variation according to response. Duration of treatment,

however, may depend upon the acuteness of the presentation. Mitchell (1966) and Norman (1970) considered that the acute or atypical actinomycosis only required a short course (1-2 weeks) of the antibiotic.

Of the various forms of penicillin, benzylpenicillin is undoubtedly the most potent. Blake (1964) reported the M.I.C. (minimum inhibitory concentration) of penicillin against strains of A. israelii to be 0.0035 - 0.015 mg/ml. This compared with phenoxymethylpenicillin, phenethicillin and ampicillin each with an M.I.C. of 0.03 mg/ml. Cloxacillin and methicillin were respectively 16 times and 128 times less effective. McGregor's (1945) pioneer treatment with direct irrigation of the lesion with penicillin proved reasonably successful but clearly the best route is by intra-muscular injection, although this is unpleasant for the patient over a prolonged period. Nevertheless, the oral route has its protagonists (Hylton et al 1970; Tyldesley 1969; Norman 1966) particularly in the treatment of the milder cases, or where children have to have protracted treatment.

Apparent lack of success with penicillin has been noted, and this has been explained in two different ways. Holm in 1951 considered penicillin effective in the eradication of the actinomyces component of the infection, but claimed that the bacillus actinomycetales comitans was resistant and could maintain the disease. He cited two cases in which the patients remained ill even after culture of the exudate failed to demonstrate actinomyces,

and concluded that the other microbes of actinomycosis appeared to have a certain conditional or relative virulence. The other explanation of the lack of success of penicillin is the resistant strain of actinomyces. Bramley & Orton (1960) noted that 4 of their 11 cases grew a species of actinomyces which was resistant in vitro to penicillin. Generally, however, actinomyces israelii is found to be sensitive to penicillin (Garrod, 1952).

Many other antibiotics have been used for actinomycosis and in this country tetracycline would appear to be the most common alternative. Bramley & Orton (1960) carried out in vitro sensitivity on their 11 cases and found tetracycline to be effective in all cases. O'Mahoney (1966) also favoured tetracycline in his treatment of 5 cases and reasoned that if the infection was mixed a wide spectrum antibiotic was more rational. Other forms of tetracycline have also been used successfully such as achromycin (Hinds & Degnan, 1955) aureomycin (McVay et al 1952) and terramycin (Lane et al 1953).

Most other common antimicrobics have been used alone or in combination. Rosh & Seldin (1948) used sulphonamide preparations but the patients also received penicillin at the same time. Erythromycin was found to be effective in vitro (Blake, 1964) and has been used clinically with success (Pizer, 1960). Isoniazid, the anti-tuberculous drug has also been utilised (McVay and Sprunt 1953, Lesney and Traeger, 1959). Isoniazid was considered

less efficient than penicillin by Baranczac (1971 in Bronner & Bronner) but was indicated in children where it was well tolerated and was administered orally. Streptomycin has also been reported used alone (Lambert, 1952) and synergistically with penicillin (Baranczac, 1971). Garrod (1952) showed that in vitro, the minimum inhibitory concentration of streptomycin was 23.7 mg/ml against penicillin with an M.I.C. of 0.06 mg/ml.

Material and Method

A retrospective review was made of the treatment of 15 cases of cervico-facial actinomycosis in Glasgow Dental Hospital and School over the past 7 years.

Details of surgical treatment and drug therapy were noted together with any complications which arose during or after treatment.

Results See Tab. 5, 1.

Discussion

In all cases, drainage of the infection was encouraged by both extraction of any associated abscessed teeth and by incision of the swelling on the external (skin) surface. (Fig. 5, 1) Where there were associated infected teeth, the duration of antibiotic therapy was relatively short (average of 3.8 weeks). In cases where the infection was chronic post traumatic in character, the antibiotic was required for a longer period which averaged 6 weeks. These findings would tend to substantiate Mitchell's (1966) view that the acute type of infection requires a less prolonged antibiotic course -

Case	Acute/Chronic	Extraction	Facial incision	Antibiotic	Duration (weeks)
1	Acute	⌈6	Yes	Penicillin	3
2	Acute	⌈6	Yes	Tetracycline	3
3	Acute	7⌉	Yes	Penicillin	3
4	Chronic	-	Yes	Penicillin	5
5	Acute	7⌉	Yes	Penicillin	-
6	Acute	7⌉	Yes	Penicillin	6
7	Chronic	-	Yes	Penicillin	5
8 (a)	Chronic	-	No	Penicillin	4
(b)	Chronic	-	No	Penicillin	9
9	Chronic	-	Yes	Penicillin Tetracycline	1 4
10	Acute	⌈5	Yes	Achromycin	3
11	Acute	-	Yes	Penicillin Erythromycin	5 2
12	Acute	-	Yes	Penicillin Erythromycin Clindamycin Cephalexin	6 5 2.5 6
13	Acute	-	Yes	Penicillin	6
14	Acute	⌈6	Yes	Penicillin Erythromycin	3 2
15	Chronic	-	Yes	Penicillin	6

Table 5, 1. Treatment of cervico-facial actinomycosis.

Fig. 5, 1. Profile view of incised actinomycotic abscess of the right cheek with short rubber drain held by a suture. (Case 13)

but only where there exists a dental focus of infection.

Recurrence in one case (Case 8) serves as a reminder that this infection is persistent and requires diligent follow-up. Penetration of the antibiotic into these fibrosed regions is thus seen to be difficult and both dosage and duration of antibiotic require constant review.

The cases in which the infection was centred on a deeper plane (Cases 11 and 12) proved most resistant to treatment. Although drainage was established in both these cases at an early stage, antibiotic therapy was more prolonged, and this was undoubtedly due to involvement of the muscle (masseter) and gland (parotid) respectively in the infection. The case involving the parotid gland was the most intransigent to treatment, and this fact re-emphasises the finding of Sazama (1965) and Hopkins (1973) that although salivary gland involvement is rare, it requires a more protracted period of anti-microbic therapy. Hopkins advocated the use of clindamycin on the grounds of its secretion through the parotid, (Stephen and Speirs, 1973) and this drug was used in the reported case. Sazama (1965) used multiple antibiotics and also used radiation in his management of 5 cases.

Penicillin and tetracyclines were the mainstay of treatment. The dosage of penicillin varied between 1 and 2 million units daily, and of tetracycline 1 Gram daily. Erythromycin was used on three occasions following penicillin therapy.

In all cases, resolution was achieved although in one case, (Case 8) recurrence resulted in scarring (Fig. 5, 2). The case which involved the parotid gland (Case 12) was, as previously noted, most unresponsive to treatment. Excessive fibrosis occurred in this case also, and limitation of mandibular movement still persists several months after resolution (Fig. 5, 3). This was caused by the binding of the skin to the underlying muscle fibres of masseter, and will require a plastic surgical procedure to separate these tissue layers. Subsequent to resolution, sialograms showed permanent parotid gland damage to be minimal.

Conclusions

1. Surgical intervention to provide adequate drainage of the abscess cavity remains fundamental to treatment, and to the establishment of a specific diagnosis.
2. Penicillin remains the drug of choice as an antibiotic with tetracycline a good alternative.
3. Dosage and duration of therapy remains a contentious issue, with each case requiring individual assessment. The success achieved by any given regime requires constant review and may require alteration of the drug employed, dosage or duration of therapy in the light of the clinical response.
4. After apparent resolution has occurred, clinical reviews are mandatory to ensure that the infection does not recur.

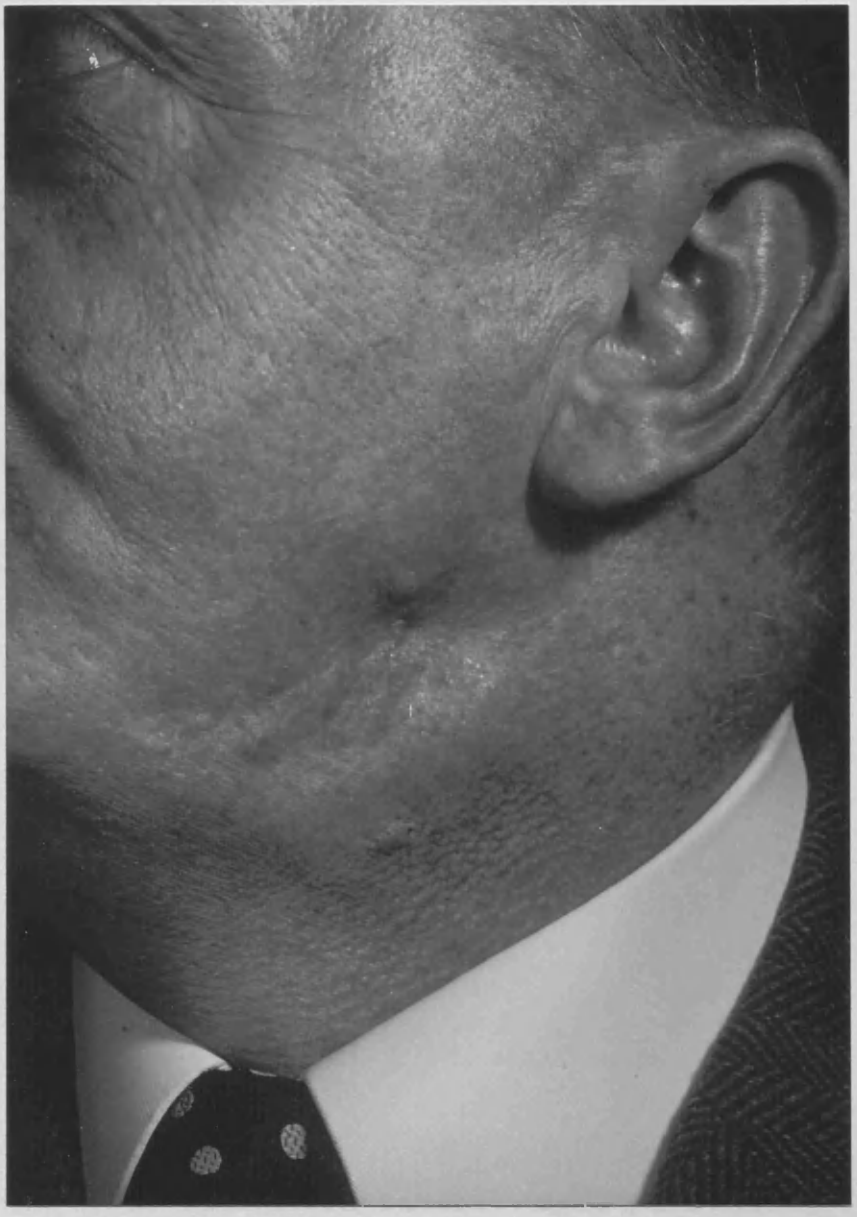

Fig. 5, 2. Severe fibrosis of cervico-facial actinomycosis resulting in scarring (Case 8).

Fig. 5, 3. Scar formation in the right parotid and masseteric area following resolution. (Case 12)

Chapter 6

Studies on the Incidence of Actinomyces in Bone Sequestra

Introduction

As has been noted in Chapter 3, actinomyces have been isolated from many intra-oral situations. Several cases in which bony sequestra were submitted to routine histopathological examination revealed organisms with the morphologic and staining characteristics of actinomyces. Perusal of the literature confirmed that infection by actinomyces was extremely uncommon in bone. (Cope, 1952; Nathan et al, 1962). However, two reports included photomicrographs of actinomycotic infection which had close similarities to these sequestra. Main & MacPhee (1964) reported a cases of actinomycosis of the maxilla in relation to a periodontal abscess, and Jurgen (1962) noted a heavily contaminated sequestrum of the mandible following jaw fracture. The possibility of necrotic bone being a source of attraction to the actinomyces micro-organisms stimulated this retrospective review, and study.

Material and Method

The records of the routine diagnostic pathology services in Glasgow Dental Hospital for the years 1968-1973 were scrutinised for all cases of bone sequestra. The morbid anatomical investigation of suspected cases of actinomycosis in the bone fragments was conducted on formalin fixed paraffin processed material. Each section was re-examined for microbes, and new sections were prepared from each block and

stained with a modified Gram stain (Brown & Brenn). Cases were diagnosed as presumptive actinomycosis if, in addition to the colonial morphology, the organisms were shown to be Gram positive branching filaments growing within tissue. Care was taken to avoid the possibility of including bacterial plaque in the specimen, and before a case was accepted as an actinomycotic infection, the tissue adjacent to the organisms had to show the characteristic acute inflammatory changes.

Relevant radiographs and clinical photographs were also traced in an attempt to evaluate the clinical features of those cases which were regarded as actinomycotic in nature. Specifically, age, sex, site and the time lapse between the extraction and sequestrum removal were sought.

Results

The sequestra were divided into two groups; a post extraction group defined as presenting within one year of an extraction at the site of occurrence, and the remainder which were classified as osteomyelitic.

Post-extraction Group

The post-extraction sequestra comprised 25 cases in total, and presence of actinomyces was found in 16 of these cases (Table 6, 1). Of the actinomycotic sequestra, 10 were in the mandible and 6 in the maxilla. This contrasted with the 9 other sequestra of which only one involved the maxilla. Comparison of the ages

using a Mann-Whitney U test of patients with actinomycotic as opposed to other sequestra revealed no significant difference, and no over-all sex difference was noted with regard to the total incidence or site involvement. The mean age of cases in the maxilla was 54.7 years as opposed to 37.7 years for cases in the mandible. This difference was highly significant using the same statistical test ($T=3.583$, $P=0.003$).

Clinically, features common to all the post-extraction sequestra included failure of the wound to heal, exuberant granulation tissue growing from the wound, (Fig. 6, 1) slight discharge, and little or no pain. Radiographically, all showed varying sizes of bony sequestrum (Fig. 6, 2 and 6, 3) lying within the socket margin and no radiolucent extension of the infection beyond the intact lamina dura.

Histological features (Fig. 6, 4 and 6, 5) other than the presence of the micro-organisms and common to all those which were found to be actinomyces positive were:-

1. The presence of necrotic bone fragments. Actinomyces were never noted in relation to vital bone.
2. No osteoclastic activity was seen in relation to the non-vital bone.
3. A polymorphonuclear leukocytic reaction in response to the organisms was noted.

4. There was a tendency for the bacteria to grow along the surface of the necrotic bone and along apparent splits in the bone.

A histogram (Table 6, 2) of the sites of this group shows maximum incidence in the lower molar and upper and lower canine areas.

Osteomyelitic Group

Osteomyelitic sequestra (Table 6, 3) were less frequent than post-extraction sequestra, and of the 10 cases, only 3 were positive for actinomyces. Such infection was thus found to be less common in cases with a more prolonged history.

Discussion

Actinomycosis of the cervico-facial region is usually associated with dental extraction in the lower molar region of the mandible - either being directly related to an abscess or following the removal of a tooth. The question arises why this area in particular is so often involved in the infection. The distribution of the sequestra obtained indicates the predominance of the lower molar region in the occurrence of such post-operative complications. These cases can be considered a random sample, since the study was essentially retrospective, and all such sequestra were scrutinized. The difficulty of extraction and denseness of the bone in the lower molar region would account for this predominance, and this is further attested by the prevalence of the canine areas which pose similar extraction problems.

Case	Age	Sex	Site	Histology H & E	Histology B & B	Time Lapse
1	37	M	⌈8	+	+	4.5 wks
2	42	M	⌈45	+	+	
3	64	M	3⌉	-	-	3 wks
4	23	M	7⌉	-	-	3 wks
5	48	F	6⌉	-	-	
6	26	M	⌈5	+	+	10 wks
7	41	F	7⌉	+	+	6 wks
8	40	F	7⌉	+	+	2.5 wks
9	53	F	⌊3	+	+	12 wks
10	32	M	4⌉	+	-	4 days
11	50	M	3⌉	+	+	4 wks
12	61	M	2⌉	+	+	24 wks
13	40	M	87⌉	+	+	
14	64	M	3⌉	+	+	4.5 wks
15	57	M	43⌉	+	+	
16	51	F	6⌉	+	-	
17	48	M	⌈7	+	+	4 wks
18	38	F	8⌉	+	+	4 wks
19	61	M	⌈6	+	-	
20	53	M	32⌉	+	+	16 wks
21	33	F	65⌉	+	-	
22	40	M	⌈3	-	-	2 days
23	48	M	7⌉	-	-	5 wks
24	27	F	8⌉	+	+	5 wks
25	25	M	⌈7	+	+	

Table 6, 1. Post-extraction group of sequestra.

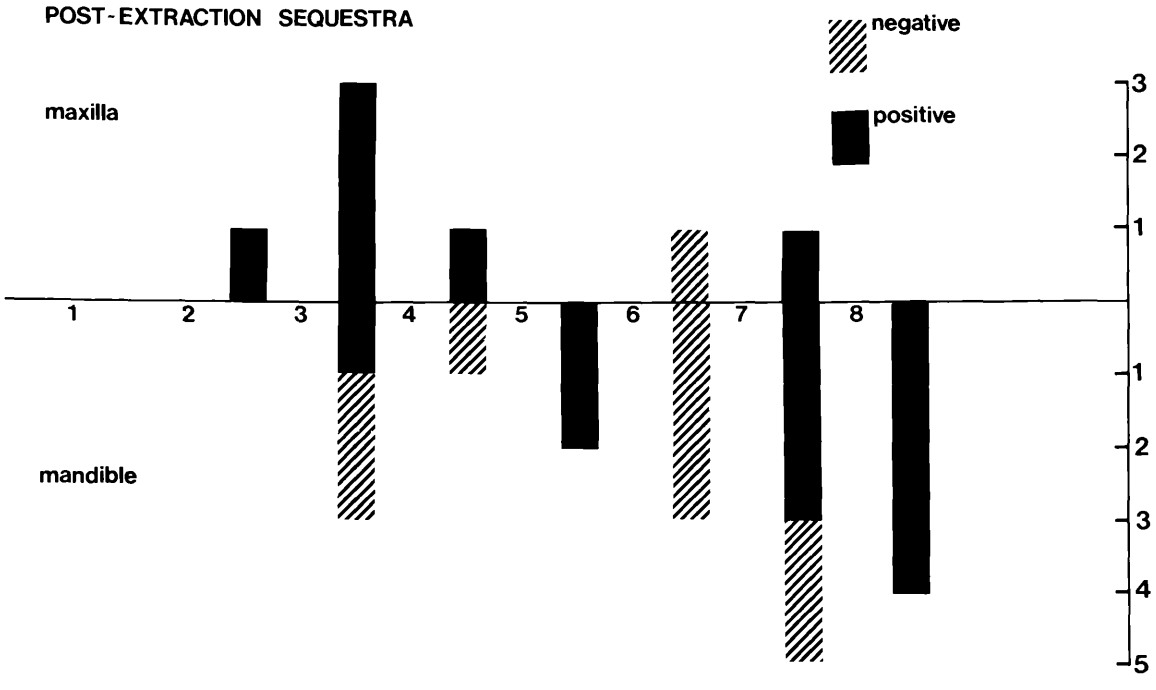

Table 6, 2. Histogram of post-extraction sequestra showing site distribution.

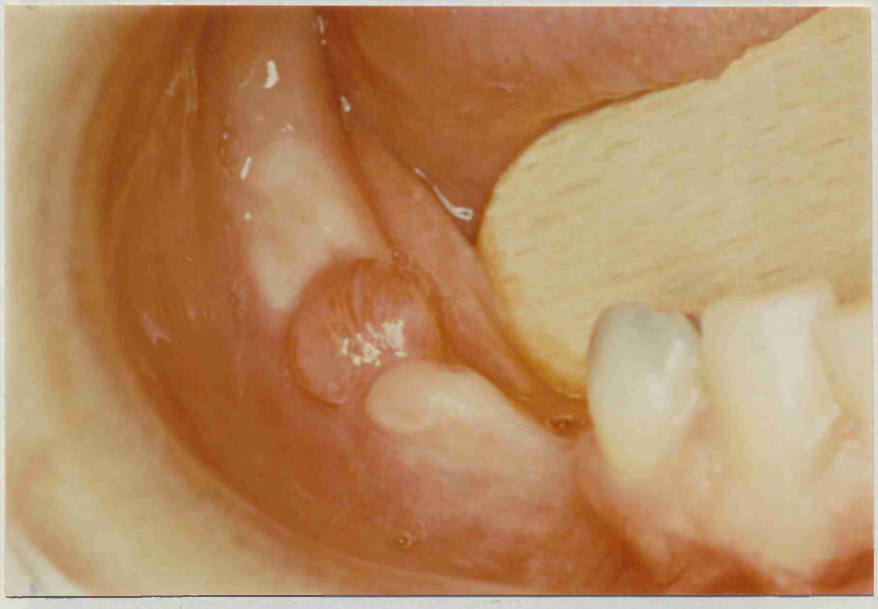

Fig. 6, 1. Clinical photograph of the typical appearance of the post extraction sequestrum. (Case II of illustrative case reports)

Fig. 6, 2. Radiograph of sequestrum lying in a lower molar socket. (Case I of illustrative case reports)

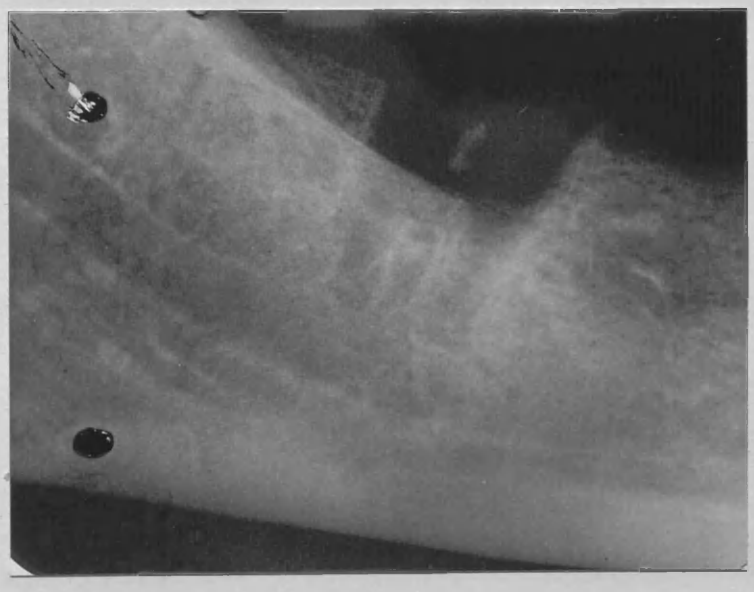

Fig. 6, 3. Radiograph of sequestrum from Case II.

Fig. 6, 4. Photomicrograph of sequestrum with gram positive filaments and bacilli in a mixed bacterial flora.
Brown and Brenn stain x 96.

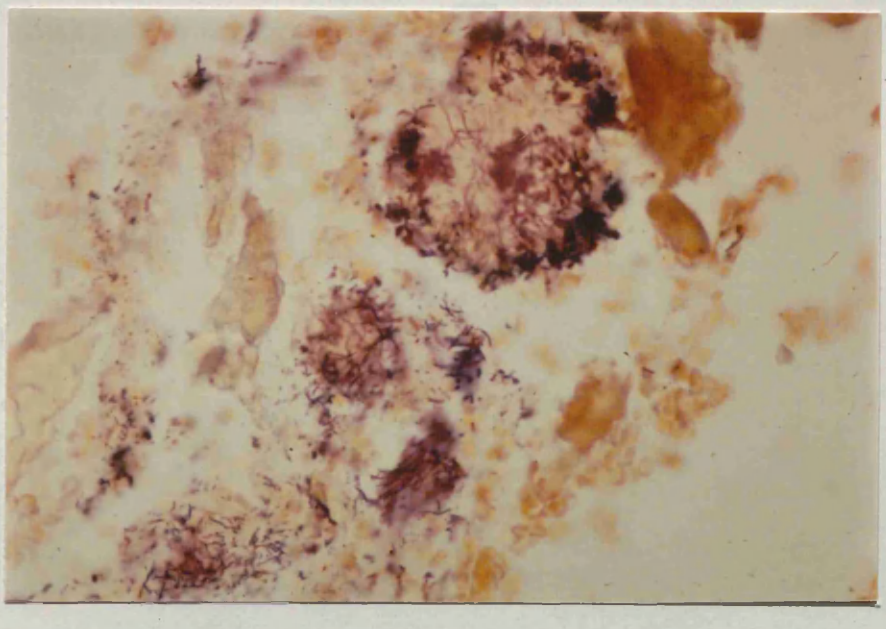

Fig. 6, 5. Photomicrograph of gram positive filaments lying within "micro" granules.
Brown and Brenn stain x 410.

Case	Age	Sex	Site	Histology H & E	Histology B & B
1	35	F	67	+	+
2	6	M	⌐	-	-
3	66	M	6	+	+
4	6	M	⌐	-	-
5	53	F	45	-	-
6	50	F	⌐	-	-
7	49	M	⌊	-	-
8	40	M	3	-	-
9	42	F	7	+	+
10	35	F	8	-	-

Table 6, 3. Osteomyelitic group of sequestra.

Whether the organisms are actively pathogenic is open to debate. It is likely, however, that the presence of actinomyces closely apposed to dead bone fragments would prevent the natural resorptive process of the body - which would occur in the absence of infection. In all the cases cited, normal healing followed surgical curettage of the area with sequestrectomy. It is possible that had these wounds remained untreated for a longer period, the infection might have invaded the adjacent soft tissues. This must be particularly true if the sequestrum has originated from the buccal or lingual alveolar plates of the extraction wound, thus opening the door to direct microbial access to the buccal or lingual soft tissues.

Summary and Conclusions

A retrospective review of sequestra from dental sources over a five year period is presented. Clinical and pathological examinations have been described with particular reference to the occurrence of actinomyces related to the bone.

The prevalence of positive findings would indicate that careful immediate post-extraction examination of sockets particularly in the lower molar region should be carried out. Any unsupported or inadequately attached bone should be removed at this time. Furthermore, the practice of leaving small sequestra to exfoliate without surgical interference in this region may be ill-advised. Thorough curettage and wound debridement

would appear to be indicated, to allow uneventful resolution of the infection, and to obviate the chance of soft tissue spread of the bacteria, and subsequent development of the classical cervico-facial actinomycosis.

Illustrative Case Reports of Post-extraction Sequestra

Case I A 48 year old male (Caucasian) presented with pain arising from the mandibular left second molar socket following an extraction 4 weeks previously. Clinical examination revealed a slight soft tissue swelling buccal to the socket, which itself appeared infected. Radiographs showed a bony sequestrum lying close to the surface of the wound (Fig. 6, 2).

Treatment consisted of sequestrectomy and curettage under local anaesthesia. Due to the acuteness of the condition, penicillin was administered by the intramuscular route prior to surgery, and the wound was packed with ribbon gauze soaked in Whitehead's Varnish (Co. Pig. Iodo.). The patient healed uneventfully.

Histopathological examination of the sequestrum revealed colonies of Gram positive filamentous organisms consistent with actinomyces.

Case II A 33 year old (Caucasian) female was referred by her dental practitioner because of a soft tissue overgrowth in the right mandible in the socket of the third molar which had been extracted one month previously. (Fig. 6, 1) On examination, there was "excessive epithelialised granulation tissue" in the socket and

radiographs showed a bony sequestrum in the wound (Fig. 6, 3).

Treatment consisted of sequestrectomy and wound debridement, following which healing progressed rapidly. Radiographs taken one year later showed normal bone regeneration in the socket.

The pathologist reported colonies of organisms morphologically consistent with actinomyces in both the connective tissue and the necrotic bone fragments.

Addendum

As a further investigation of bone sequestra infected with actinomyces, an in vitro experiment was carried out for the purpose of ascertaining the affinity of the micro-organism to necrotic bone fragments.

Material and Method

Bone fragments were obtained from human lower third molar areas during surgical removal of impacted lower wisdom teeth. The fragments were washed thoroughly in saline, and then sterilised by autoclaving. The sterilised fragments were then suspended in test tubes containing Brewers Thioglycollate Broth, and a pure culture of actinomyces israelii, actinomyces naeslundii and an anaerobic streptococcus species was innoculated into the three prepared test tubes. One fragment was left uninnoculated as a control.

The test tubes were then left at 37°C for a period of 7 days. After this incubation, the bone fragments were removed, fixed in formalin 10% solution,

Fig. 6, 6. Photomicrograph of actinomyces israelii forming along bone fragment surfaces in vitro. Compare with Fig. 6, 4. Brown and Brenn x 96.

decalcified and mounted in paraffin wax. Sections were then cut and stained with haematoxylin and eosin and with Brown and Brenn (modified Gram) stain.

Results

Both species of actinomyces showed an affinity to grow around the fragments of necrotic bone. The anaerobic streptococcus also grew along the bone surface, but retained the coccal morphology. Comparison of the sections obtained from clinical sequestra (Fig. 6, 4) and the in vitro experimental sections (Fig. 6, 6) show similarities. In particular, the tendency of the filaments to align along the surface of the bone and within the splits was duplicated in the in vitro situation.

Conclusions

Since there was no attempt to innoculate the bone fragments directly with the bacteria, there does appear to be an attraction to non-vital bone. The tendency of the bacteria to grow along the surface of the bone could explain the lack of resorption in the clinical cases described previously.

Chapter 7

Investigation into the Prevalence of Actinomyces Israelii and Actinomyces Naeslundii in Human Dental Plaque

Introduction and review of literature

Actinomyces are common micro-organisms in the oral and pharyngeal regions. They have been successfully isolated from such sites as tonsillar crypt (Emmons, 1938) dental plaque (Morris, 1954; Bibby & Knighton, 1941; Ennever et al, 1951; Howell et al, 1962; Snyder et al, 1967) and dental calculus (Slack et al, 1971; Collins et al, 1973). These studies have shown a progressive development of technique in the indentification of the actinomyces from morphology and staining reactions, colonial characteristics and biochemical reactions, to fluorescent antibody testing using species specific antiserum. (Slack et al 1960). Of the several strains which can be identified, A. israelii is the most common infecting agent in man although A. naeslundii has also been reported in a pathogenic role (Coleman et al, 1969; Coleman and George, 1969; Socransky et al, 1970; Jordan et al, 1972). A. israelii is a strict anaerobe whilst A. naeslundii is an aerobe and facultative anaerobe (Thompson & Lovestedt, 1951) and this is probably the most important fundamental difference between the two species, although biochemically they also display differences in sugar fermentation reactions (Slack et al, 1969, Pine et al, 1960). (Tab. 7, 1)

Attention has been drawn in Chapter 3 to the striking prevalence of the lower molar region in cases of cervico-facial actinomycosis and in Chapter 6, the inference between difficulty of extraction in this area

and occurrence of actinomyces containing sequestra was drawn. Logically, the prevalence of the micro-organisms would appear to be pertinent in different areas of the oral cavity. In most of the previous studies referred to above, little attention has been drawn to the exact site of procurement of the sample, whether of dento-bacterial plaque or dental calculus. Clearly, the bacterial population of plaque samples must vary according to the exact site in the oral cavity which it occupies, and material from the anterior labial region has environmental differences such as oxygen availability and salivary influence from material obtained from the posterior regions of the mouth. (Eskow & Loesche, 1971)

The following study was carried out to examine the possible differences in the prevalence of A. israelii and A. naeslundii in dento-bacterial plaque from the anterior maxillary labial region and the posterior mandibular lingual region in given individuals.

Materials and Method

Dental plaque was collected from random individual patients attending Glasgow Dental Hospital for a variety of conditions - mainly casual extractions. In each case, plaque was collected from two areas - namely the upper anterior labial surfaces of the canine to canine region and the lower molar lingual region of either side. The plaque was taken in each case from the cervical one third of the clinical crown supra-gingivally. The samples were

taken with a sterile dental plastic instrument, and transferred to a standard sterile micro-loop (A/S Nunc. Denmark) (Fig. 7, 1) from which excess was carefully removed.

Each loop was innoculated in 2 ml. of phosphate buffer saline and mixed for at least 2 minutes by mechanical agitation (Whirlimixer, Fisons Scientific Apparatus Loughborough). After through dispersal, a standard loop of each sample was smeared on a clean glass slide and allowed to dry. Each smear was made as close to 1 cm. in diameter as possible. After drying, the smears were gently heat fixed. Two such smears were prepared for each sample.

Fluorescent antibody test. The fluorescent antiserum was obtained from the Biological Reagents Section, National Center for Disease Control, Atlanta. Two sera were employed; A. israelii conjugate of serotypes 1 and 2, and A. naeslundii (absorbed with A. israelii cells). The techniques for testing were identical for all smears and antisera.

The fluorescent antiserum was layered onto the prepared smear then placed in a moist chamber for 30 minutes, after which thorough washing with fluorescent antibody buffer (Bacto. fluorescent antibody buffer) was carried out. The slides were mounted on buffered glycerol (9 part buffer to 1 part glycerol) and examined with a Wild M20 microscope illuminated by an HBO 200W. mercury lamp and using an F.I.T.C. barrier filter in conjunction with a B.G.12 filter.

	Growth in air	Catalase	Glucose	Xylose	Cellobiose	Raffinose
A. israelii	-	-	Acid	Acid	usually Acid	Acid
A. naeslundii	+	-	Acid	-	-	usually Acid
A. odontolyticus	+	-	Acid	-	variable	-

Table 7, 1. Oxygen requirements and biochemical reactions of actinomyces species.

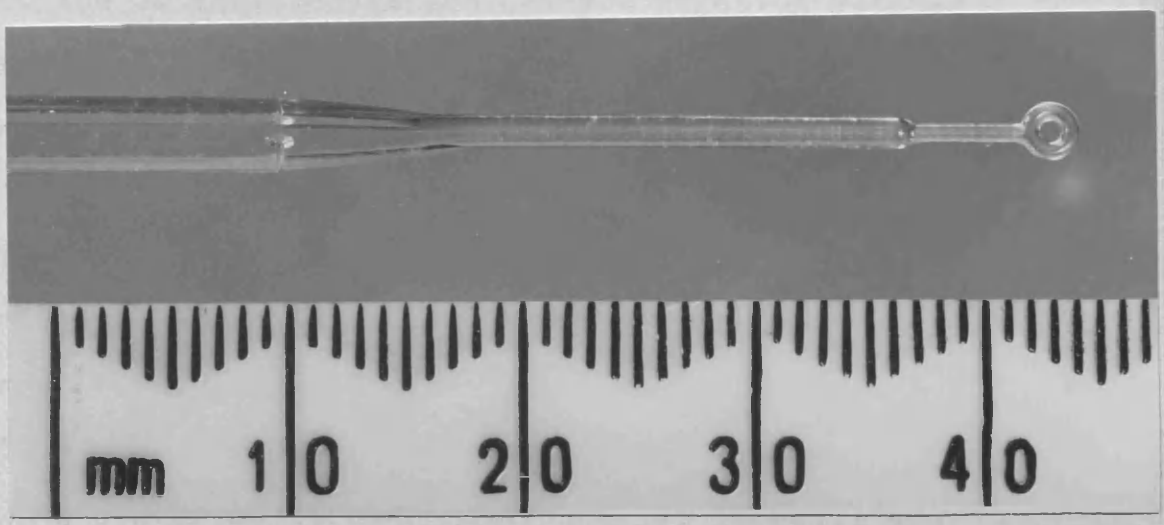

Fig. 7, 1. A/S Nunc sterile micro-loop.

Each preparation was examined under x20 magnification and fluorescent bacteria were counted. For each preparation, 3 random fields were counted within the smear by two assessors. For each field, the assessor's counts were averaged, and a mean of the 3 fields was then obtained.

Results

A total of 20 individuals contributed to the study of A. israelii of whom 16 were also examined with regard to A. naeslundii. The age range for the A. israelii group was 16 to 57 with an average of 29 years and for the A. israelii/A. naeslundii group the ages ranged similarly with an average of 30.1 years.

From table 7, 2 it can be seen that the posterior A. israelii count was greater than the anterior count in 17 of the 20 individuals and using a Wilcoxon Matched Pairs Signed Rank Test ($T=28$ $P< 0.01$) this was highly significant.

Similarly from table 7, 3, using the same statistical test, it was found that there was no significant difference in the A. naeslundii counts in the anterior and posterior regions for any given individual ($T=57.5$ $P>0.05$)

A comparison of the numbers of A. israelii and A. naeslundii in the posterior region (Table 7, 4) showed that A. israelii gave a higher count than A. naeslundii in 14 of the 16 individuals. This again was found to be highly significant using a Wilcoxon Matched Pairs Signed

Rank Test ($T=17$ $P<0.01$). In the anterior region (Tab. 7, 5) A. israelii was found to have a higher count than A. naeslundii in 11 of the 16 cases which was barely significant ($T=30$ $P=0.05$).

The total A. israelii count (anterior and posterior) compared with the total A. naeslundii count (Table 7, 6) showed A. israelii to be significantly more prevalent than A. naeslundii ($T=27$ $P<0.05$).

Discussion and Conclusions

Most quantitative counts of bacterial populations in dental plaque or calculus have involved investigation of a standard weight of the material. In this study, a standard volume was examined, and no attempt was made to assess the total or viable bacterial population. In strict micro-biological terms, therefore, the counts of bacteria in this investigation cannot be directly compared with other studies in which such enumeration was carried out (Collins et al, 1973; Snyder et al, 1967). The main purpose of this work was to compare the occurrence of A. israelii and A. naeslundii in a standard volumetric quantity of plaque taken from differenct environments in a given individual. Nevertheless, on a volumetric basis, some information on the relative prevalence of these two micro-organisms is gained.

The results show that A. israelii is present in significantly larger numbers in the posterior lingual region than in the anterior labial region, whereas no such difference is true of A. naeslundii. The strict

anaerobic requirements of A. israelii would explain this finding, since the posterior lower quadrants do provide an environment more conducive to anaerobic growth. The lingual aspect of the lower molars in particular would appear most suitable, since the tongue overlaps this area and it forms a natural drainage area in the mouth. A. naeslundii being less demanding of anaerobic conditions appears to exist equally well in both sites.

Oxygen availability would also explain the relatively higher numbers of A. israelii than A. naeslundii in the posterior samples, though the result in this comparison must be qualified by a similar finding anteriorly. This would suggest that A. israelii is a more prevalent organism generally in dental plaque, and this indication is substantiated by recent work of Bowden et al (1975) who showed that the dominant species of actinomyces in interproximal plaque was A. israelii with a mean percentage of 16.5 compared with A. naeslundii and A. viscosus with a combined percentage of 19.05. The results obtained here also indicate that the total count of A. israelii is significantly greater than the A. naeslundii count regardless of site. Bowden's paper (1975) also indicated that the genus Actinomycetales was predominant in dental plaque with a mean percentage of 34.92 compared to streptococcus, the next most prevalent with a mean percentage of 22.9.

The direct examination of the specimens supports the

finding of Snyder et al (1967) that both A. israelii and A. naeslundii exist in vivo as short bacillary or coccal forms rather than the filamentous form found most frequently in pathological material from actinomycotic infection. Sims (1974) considered that this filamentous morphology accounted for its persistence in the tissues by rendering it more resistant to phagocytosis.

Clinically, there is a well recognized post traumatic group of cases of actinomycosis with prevalence of the lower molar region. Chapter 6 has indicated that difficulty of extraction in this area could account for an increased incidence of sequestrum formation with contamination with actinomyces. From this investigation, it emerges that in this lower molar area there is an increased population of A. israelii within the plaque, and this must increase the chance of access to any wound in this region, and ultimately to the development of soft tissue infection and the typical cervico-facial actinomycosis.

Case	Age	Sex	Anterior	Rank	Posterior	Rank
1	16	M	51	14.5	22	29
2	27	M	60	13	82	-22
3	16	F	32	16.5	66	-34
4	38	M	51	11	68	-17
5	39	M	173	5.5	184	-11
6	36	M	28	19	85	-57
7	31	M	23	1.5	27	- 4
8	25	M	48	8	36	12
9	25	M	19	1.5	23	- 4
10	26	M	66	18	116	-50
11	32	M	51	14.5	80	-29
12	27	M	21	8	33	-12
13	23	M	15	20	73	-58
14	48	M	65	16.5	99	-34
15	57	M	24	8	36	-12
16	16	M	34	3	43	- 9
17	23	M	27	4	37	-10
18	19	M	40	5.5	29	11
19	35	M	13	11	30	-17
20	22	M	34	11	51	-17

Table 7, 2. A. israelii (anterior)
Vs. A. israelii (posterior)
Wilcoxon Matched Pairs Signed Rank Test.
T=28 $P < 0.01$.

Case	Age	Sex	Anterior	Rank	Posterior	Rank
1	16	M	6	8	15	- 9
2	27	M	13	5.5	19	- 6
3	16	F	28	5.5	34	- 6
4	38	M	39	2	37	2
5	39	M	50	12	20	30
6	36	M	3	10	16	- 13
7	31	M	23	10	10	13
8	25	M	21	3.5	24	- 3
9	25	M	21	3.5	18	3
10	26	M	38	14	95	- 57
11	32	M	102	15	346	-244
12	27	M	85	13	34	51
13	23	M	16	1	17	- 1
14	48	M	24	10	11	13
15	57	M	10	7	2	8
16	16	M	17	0	17	0

Table 7, 3. A. naeslundii (anterior)
Vs. A. naeslundii (posterior)
Wilcoxon Matched Pairs Signed Rank Test
$T=57.5 \quad P > 0.05$.

Case	Age	Sex	A. Israelii	A Naeslundii	Difference in count d	Rank of d
1	16	M	22	15	7	3
2	27	M	82	19	63	12
3	16	F	66	34	32	9
4	38	M	68	37	31	8
5	39	M	184	20	164	15
6	36	M	85	16	69	13
7	31	M	27	10	17	5
8	25	M	36	24	12	4
9	25	M	23	18	5	2
10	26	M	116	95	21	6
11	32	M	80	346	-266	-16
12	27	M	33	34	- 1	- 1
13	23	M	73	17	56	11
14	48	M	99	11	88	14
15	57	M	36	2	34	10
16	16	M	43	17	26	7

Table 7, 4. A. israelii (posterior)
Vs. A. naeslundii (posterior)
Wilcoxon Matched Pairs Signed Rank Test.
T=17 $P < 0.01$.

Case	Age	Sex	A Israelii	A Naeslundii	Difference in count d	Rank of d
1	16	M	51	6	45	11
2	27	M	60	13	47	12
3	16	F	32	28	4	3
4	38	M	51	39	12	4
5	39	M	173	50	123	15
6	36	M	28	3	25	7
7	31	M	23	23	0	
8	25	M	48	21	27	8
9	25	M	19	21	- 2	- 2
10	26	M	66	38	28	9
11	32	M	51	102	- 51	-13
12	27	M	21	85	- 64	-14
13	23	M	15	16	- 1	- 1
14	48	M	65	24	41	10
15	57	M	24	10	14	5
16	16	M	34	17	17	6

Table 7, 5. A. israelii (anterior)
Vs. A. naeslundii (anterior)
Wilcoxon Matched Pairs Signed Rank Test.
T=30 P=0.05.

Case	Age	Sex	A Israelii Anterior Posterior	Rank	A Naeslundii Anterior Posterior	Rank
1	16	M	73	9	21	52
2	27	M	142	13	32	110
3	16	F	98	3	62	36
4	38	M	119	5.5	76	43
5	39	M	357	15	70	287
6	36	M	113	12	19	94
7	31	M	50	2	33	17
8	25	M	84	4	45	39
9	25	M	42	1	39	3
10	26	M	182	8	133	49
11	32	M	131	16	448	-317
12	27	M	54	11	119	- 65
13	23	M	88	10	33	55
14	48	M	164	14	35	129
15	57	M	60	7	12	48
16	16	M	77	5.5	34	43

Table 7, 6. A. israelii (anterior + posterior) Vs. A. naeslundii (anterior + posterior) Wilcoxon Matched Pairs Signed Rank Test.
$T=27$ $P < 0.05$.

Chapter 8

Actinomyces in the Edentulous Subject

Introduction

Actinomycosis in the cervico-facial region is primarily a disease associated with the dentate subject. In the vast majority of reported cases the infection can be traced to a dental source, either an apical dental abscess or an extraction wound or jaw fracture. However, a few cases do not seem to have any direct dental link. Holmes (1958) reported a case of cervical actinomycosis with no dental focus or wound which later spread to the lung and culminated in the patient's death. Stanton (1966) reported a case which involved both maxillary antra in a female of 52 years who had been edentulous for a period of 17 years and who had undergone tonsillectomy at the age of 15 years. In the same paper, he discussed another case which involved the antrum with no dental focus apparent first described by Harsch in 1945. Sazama (1965) reported one case of parotid actinomycosis with no direct dental involvement, but the presence or otherwise of teeth was not mentioned. Hopkins (1973) also reported a case of primary actinomycosis of the parotid gland though the patient was dentate, as was the patient treated in this institution with salivary gland infection (Case 12). In another case (Case 13) a partial upper denture may have been the agent which, by causing mucosal ulceration, allowed ingress of actinomyces into the soft tissue of the cheek, but here again, the patient was dentate, albeit partially.

A paper by Bowden et al (1975) indicated the

prevalence of actinomyces in dental plaque which constituted a mean percentage of 34.92 compared with streptococcus with a mean percentage of 22.9 the next most prevalent organism. This paper also showed that within the genus, A. israelii was the predominant species of actinomyces within dental plaque with a mean percentage of 16.5. Clearly, actinomyces abound within the oral microbial flora of the dentate subject.

The relationship of the actinomyces to the eruption of the teeth was reported by McCarthy et al (1965). This investigation showed that immediately after birth actinomyces were not present in the oral cavity of the 51 cases studied. During the first year of life, however, they became progressively more prevalent, such that at 1 year the organism was recovered from 17 of 29 cases. During the first year of life, 12 deciduous teeth normally erupt, and McCarthy et al thus showed the close dependence of actinomyces on the presence of teeth.

No information, however, has been reported of the existence of actinomyces in the edentulous subject, and an investigation was therefore instituted to clarify the effect of total loss of the dental tissues on the presence of these micro-organisms.

Materials and Method

Debris was collected from the fitting surface of complete upper dentures with sterile dental plastic instruments. These dentures were of varying age, and the

subjects had been edentulous for a varying period which was recorded. A standard volume of the scraping (A/S Nunc micro-loop) was transferred to 1 ml. phosphate buffer saline, and thorough dispersal was achieved by mechanical agitation using a Whirlimixer. (Fisons Scientific Apparatus, Loughborough.) Serial dilutions were then prepared from 10^{-1} to 10^{-5}, and these dilutions were then innoculated (1 standard loop, 0.1 ml.) onto blood agar plated, and evenly spread over the entire surface. The plates were then incubated anaerobically (10% CO_2 90% H_2) at 37°C for 7 days.

Colonies were then examined for the typical crenated surface and adherence to blood agar which is characteristic of A. israelii. Suspected colonies were tested with Gram stain, and if they showed Gram positive bacilli or filaments, then a direct fluorescent test was carried out as described in Chapter 4 using both A. israelii antiserum and A. naeslundii antiserum as control.

Results

Details of the age and sex of each subject together with the length of time edentulous in terms of a period greater or less than 1 year are shown in Tab. 8, 1. A dilution of 10^{-3} was found to achieve the best separation of colonies for assessment. Approximate quantification of the organisms present was made on the basis of a light growth (0-10 colonies), moderate growth (10-40 colonies) and heavy growth (>40 colonies).

In only one case was A. israelii isolated from a

Cultures of Fitting Surface from Full Upper Dentures

Case	Sex	Age	Edentulous over 1 year	Edentulous less than 1 year	Culture 10-3
1	F	60	+		Streptococcus viridans +
2	F	48		6 months	Streptococcus viridans +
3	M	67	+		Streptococcus viridans ++
4	F	40		2 months	Streptococcus viridans +++ Anaerobic streptococcus ++ Bacteroides melaninogenicus Vibrio species + Fusobacterium +
5	M	51	+		No Growth
6	F	52	+		Streptococcus viridans ++
7	F	54	+		Streptococcus viridans + Anaerobic streptococcus +
8	F	28		1 month	Streptococcus viridans +++ β Haemolytic streptococcus + Fusobacterium + Eikenella Corrodens + Actinomyces israelii +
9	M	68	+		Streptococcus viridans +
10	F	77	+		Streptococcus viridans ++
11	M	47		4 months	No Growth
12	F	42	+		No Growth
13	M	69	+		Streptococcus viridans +

Table 8, 1.

total of 13 subjects. In 3 samples, no growth was obtained and in 10 samples, streptococcus viridans was grown. This organism was found alone in 7 cases, and was the most prevalent in those cases which yielded a mixed bacterial flora. Two samples were found to grow a flora consisting of 5 separate species. In both these cases streptococcus viridans grew heavily.

Discussion and Conclusions

Actinomyces israelii was recovered from the subject who had been rendered edentulous most recently, and from this sample and the other most recently dentate (1 and 2 months respectively) a flora consisting of 5 species was grown. These flora most closely resemble the type of admixture of organisms which the normal dental plaque would yield. In all the cases edentulous for more than 1 year, a sparse growth was obtained devoid of actinomyces.

It would appear that loss of the natural dentition is followed by a progressive reduction in the Actinomyces israelii population at least from a site which the anaerobic requirement of the bacterium would almost certainly favour. Clearly, however, a longditudinal study on the effect of total dental extractions would reveal whether a gradual diminution of A. israelii numbers paralleled the emergence of the species shown by Snyder et al (1967). The presence of just one tooth or root, nevertheless, is enough to harbour A. israelii within the mouth, and indeed to produce an actinomycotic infection (Case 6 and 10).

Chapter 9

Experimental Phagocytosis of Actinomyces by Human Polymorphonuclear Leucocytes

Introduction

Cervico-facial actinomycosis is a disease characterised by a marked fibrotic reaction in the host tissue. This fibrosis has been illustrated in Fig. 5, 2 and 5, 3 and can cause permanent disfigurement. Actinomyces israelii is susceptible to most antibiotics (Garrod, 1952, Blake, 1963, 1964) and in particular to penicillin. It is not easy therefore to understand why resolution is so often protracted, and hence treatment is also of prolonged duration. Sims (1974) offered several suggestions as to why this should be so, amongst which was the theory that the organisms were not easily phagocytosed by the polymorphonuclear leucocytes. He further surmised that lysosomes released by these "thwarted" phagocytes could be responsible for the tissue destruction of the disease process. A factor such as the filamentous morphology of the microbe could, he argued, be responsible for the initial difficulty in the process of phagocytosis. Hence an investigation was undertaken to assess the ability of polymorphonuclear leucocytes to ingest known actinomyces micro-organisms in vitro.

Material and Method

The technique used was advocated by Wright and Colebrook (1921) for the study of the emigration of leucocytes.

A cell or round compartment was delineated on a clean glass slide using paraffin wax. This cell of approximately 1 cm. in diameter was filled with a heaped

up drop of blood obtained from a healthy male adult, and the slide was incubated at 37°C for ½ hour. Moisture was maintained in the sample by placing it in a Petri dish lined with blotting paper soaked in warm water.

After incubation, the clot was shaken off the sample with a sharp downward jerk of the wrist, and any residual red cells were carefully washed off the sample by gentle irrigation with tissue culture fluid. (Parker's 199 tissue culture medium; Flow Laboratories, Irvine, Scotland). The leucocytes remain as a single layer of cells visible as a grey film over the glass. The wax was then removed using a sharp scalpel.

A known species of Actinomyces israelii (N.C.T.C. 10215), Actinomyces naeslundii (N.C.T.C. 10301) and an an anaerobic streptococcus were each incubated in tissue culture medium at 37°C for 30 minutes and several drops of each was pipetted onto the layer of leucocytes on 3 slides. One slide did not receive an innoculum and was kept as a control. Each preparation was covered by a clean dry cover slip and the cover slip was sealed with paraffin wax around its periphery to prevent excessive drying. Incubation was then carried out and progress of the phagocytic process was scrutinised at regular intervals using phase contrast microscopy on a preheated (37°C) stage. (Leitz Ortholux with phase attachment). Each slide was incubated in a moist Petri dish.

After ¾ hour, the slides were removed from the incubation chamber, and the cover slips were carefully removed. The preparations were fixed in acetone, stained by the Leishmann method, and viewed under light microscope.

The experiment was carried out on two occasions.

Results Quantitation of the phagocytic activity was not attempted, but there appeared to be no impediment to ingestion of any of the test organisms. Bacteria of both cocco-bacillary and filamentous forms of actinomyces were ingested, and up to 15 microbes were seen in one phagocyte. (Figs. 9, 1; 9, 2; 9, 3)

Discussion and conclusions

The tissue damage which results from actinomycosis can be gross and result in deformity. Since no exogenous toxins from actinomyces have been isolated or described, the cause can be reasonably assumed to be either from concomitant bacteria where the infection is mixed, or from lysosomes released from the breakdown of the leucocyte. Most bacteria are broken down rapidly within the white cell but some, such as mycobacteria tuberculosis and brucella species survive such engulfment. Death of most micro-organisms occurs rapidly, usually within 15 minutes for streptococci (Hirsch, 1974). In terms of a morphological impediment to phagocytosis, there would appear from this study to be none - at least in vitro. Actinomyces have, however, an ability to deposit calcium salts intra-cellularly (Ennever 1960) and the sulphur

granule seen in pathological specimens has been estimated to contain 50% calcium phosphate in a polysaccharide-protein matrix (Pine and Overman, 1963). This calcified matrix may well form a protective barrier to the polymorph which will physically prevent the cell contacting the microbe. The polymorph may then release the lysosomal enzymes without being actively damaged as part of a phenomenon known as "frustrated phagocystosis." (Hirschhorn 1974). Formation of sulphur granules is a common finding in the active disease, (Chap. 3 and 4) and the tissue destruction may well be due to rupture of effete bacteria-laden phagocytes and the enzymes released from the frustrated cells. Additional difficulty in resolution of the infection is the fibrotic reaction of the host tissue which must impede not only antibiotic permeation into the affected region, but also transport of exudate both to and from the abscess cavities. The more protracted antibiotic course required for the chronic form of the disease would tend to support this contention since the tissues have a longer period to "wall off" the infection with fibrous tissue.

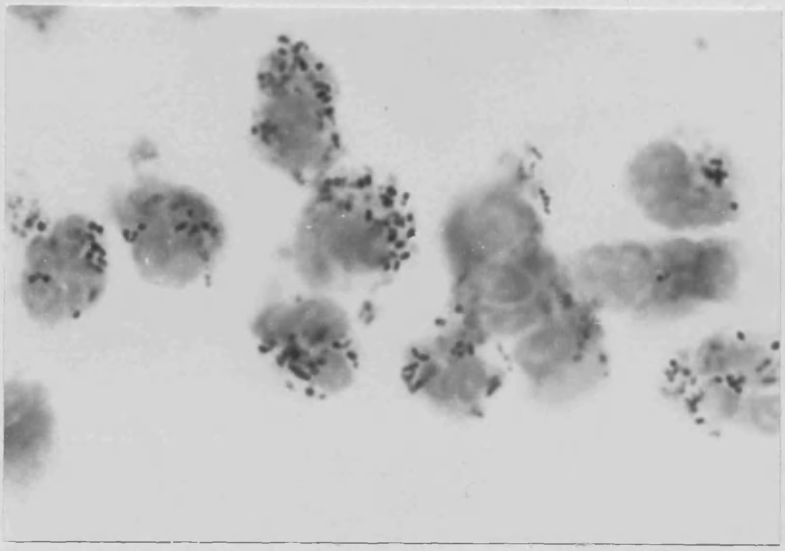

Fig. 9, 1. Photomicrograph of actinomyces israelii species lying within polymorphonuclear leucocytes.
Leishmann stain x 960.

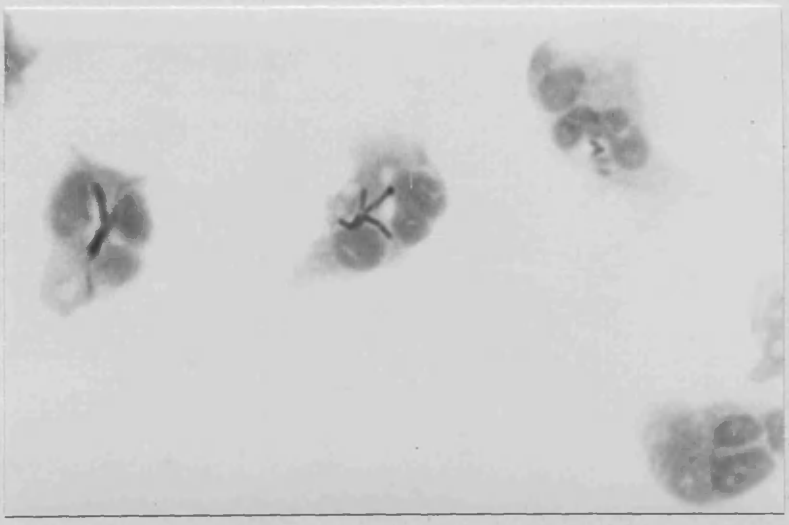

Fig. 9, 2. Photomicrograph of filamentous forms of actinomyces naeslundii after phagocytosis.
Leishmann stain x 960.

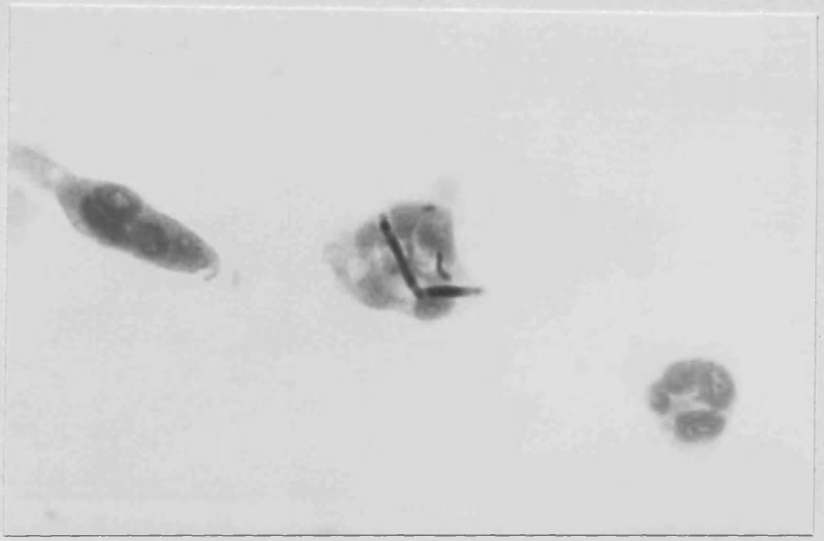

Fig. 9, 3. Photomicrograph of a large filament of actinomyces naeslundii within the phagocyte which also has engulfed another shorter bacillary form.
Leishmann stain x 960.

Appendix A

Precis of Case Reports of Cervico-facial Actinomycosis

Case 1

An 11 year old female Caucasian attended the Glasgow Dental Hospital with a swelling in the left side of the mandible of 5 days duration. Pain was moderate.

On examination, the swelling was tender to palpation and there was a related trismus and submandibular lymphadenitis. Intra-orally a grossly carious /6 was noted and radiographs confirmed that periapical infection was present.

Under general anaesthesia, the /6 was extracted and the external soft tissue swelling was incised and drained. Bacteriological examination of the pus showed the infection to be actinomycosis, and a total of 3 weeks penicillin orally resulted in resolution.

Case 2

(Female Caucasian aged 13) This patient complained of a swelling on the left side of the lower jaw which had arisen 6 weeks previously but had only become acute within the last week. An inadequate penicillin course of 2 days had been given by her prectitioner but she had failed to return to him for further treatment.

On examination, an acute left submandibular abscess was noted and a carious /6. Periapical involvement of this tooth was confirmed radiographically.

Under general anaesthesia, the facial abscess was incised and the /6 extracted. Pathological examination of the curettings showed typical actinomycotic granules

although bacteriological culture failed to isolate the bacterium due to overgrowth by other micro-organisms. The patient was treated with a total of 3 weeks tetracycline orally and resolution was unimpeded.

Case 3

A 17 year old male Caucasian and a chef by profession, complained of pain and a swelling of the right lower jaw. This swelling had been present for 3 weeks and had been treated with penicillin injections over the last week by his medical practitioner.

On examination, he showed a large (3" diameter) fluctuant swelling in the right submandibular area, and a related carious $\overline{7}/$. This tooth was extracted and the abscess incised and drained. Bacteriological examination of the discharge revealed the infection to be actinomycotic and the patient had a total of 3 weeks penicillin.

Case 4

A 27 year old male Caucasian and "fitter" by trade. The patient presented with a history of pain in the lower left jaw which had appeared following dental extractions including $/\overline{7}$ some 7 weeks previously.

On examination, a tender diffuse swelling related to the left body of the mandible was noted. Intra-orally, there was no evidence of retained roots, and a diagnosis of periostitis was made. Radiographs showed nothing abnormal.

The patient was put on penicillin therapy for 1 week and then erythromycin for 1 week since resolution had not

occurred. For the ensuing 6 weeks, the swelling slowly migrated to the submandibular region where it eventually presented as a "button" of erythematous tissue and was incised and drained. A swelling anterior to this was noted at this time in the left submental region which eventually pointed to the surface. This latter swelling was also incised and the pus was reported as actinomycotic. A total of 5 weeks penicillin therapy achieved good resolution.

Case 5

A 26 year old female Caucasian housewife. This patient attended complaining of severe throbbing pain in the right lower jaw. She had first noticed the swelling 4 weeks previously and had been treated with penicillin, which had had little beneficial effect.

Examination revealed a large swelling related to the right angle of the mandible and submandibular region. There was a marked trismus but roots in the $\overline{7}7$ area were noted and confirmed radiographically.

Treatment consisted of incision and drainage of the swelling and extraction of $\overline{7}7$ roots. Pathological examination of the exudate allowed the diagnosis of actinomycosis and the patient was put on a course of oral penicillin. She failed to keep repeated appointments, and a letter was sent to her medical practitioner requesting him to continue the penicillin.

Case 6

(20 year old Caucasian female factory worker) The patient was referred to the Glasgow Dental Hospital with

a hard painful swelling in the right mandible. It had first arisen 5 weeks previously and had subsided following penicillin administration from the medical practitioner.

Clinical examination revealed a very hard fibrotic submandibular swelling on the right side with retained $\overline{7}7$ root in an otherwise edentulous mouth.

Treatment consisted of external incision and drainage, removal of the root $\overline{7}7$, and a six weeks course of phenoxymethylpenicillin orally, following the bacteriological report of actinomycosis.

Case 7

(44 year old female Caucasian housewife) The patient was referred with a painless hard lump associated with the body of the right mandible. It had arisen 4 weeks after extraction of $\overline{8}7$ and had been treated with little success with a 1 week course of penicillin prior to seeking advice at Glasgow Dental Hospital.

Examination clinically revealed a hard relatively painless mass about $\frac{1}{2}$ inch in diameter and firmly attached to the bone. It was variously diagnosed as periostitis, an aberrant lymph node adenitis, and soft tissue abscess, and over a period of 4 weeks it eventually softened and presented on the skin surface. It was incised and drained under general anaesthesia but at this time another swelling was noted anterior to the original lump. Bacteriological examination of this incision revealed a mixed infection which included a Gram positive filament. One week later,

the anterior swelling came to the surface and was incised. Actinomycosis was confirmed with this discharge, and the patient was treated with a 5 weeks penicillin course. Resolution was achieved uneventfully.

Case 8

Caucasian male aged 46 (factory worker). This patient was referred by his dentist with swelling in the left submandibular region which had been present for 7 weeks. His dentist had removed roots /78 area during this time with no effect on the swelling.

On examination, multiple sinuses were seen in the left submandibular region associated with a diffuse swelling which showed much induration and fixation of skin. There was trismus and some swelling in the buccal sulcus in the molar area. The discharge was noted to contain a sulphur granule by the clinician.

Following bacteriological confirmation of actinomycosis, the patient received 4 weeks intra-muscular penicillin to which he responded well. He was reviewed regularly and 3 months after his initial attendance at Glasgow Dental Hospital and School, he was given a check-up appointment for 6 months time.

At this appointment, the patient reported intermittent swelling and discharge over the 6 month period which he had not thought fit to report. A recurrence was diagnosed although there was no active sinus present at the time. A further 9 weeks oral penicillin was prescribed, and resolution was achieved.

Case 9

29 Year old Caucasian housewife. The patient presented with a hard fixed swelling related to the right body of the mandible. She had a history of surgical removal of $\overline{8}/$, 12 weeks prior to the swelling appearing. A week after presentation this swelling reddened on the skin surface and was incised and drained.

Following the bacteriological report of actinomycosis, the patient was given a 1 week course of penicillin which caused some gastro-intestinal upset, and the antibiotic therapy was therefore continued with 4 weeks tetracycline. Resolution was uneventful.

Case 10

40 year old Caucasian female (assembler) The patient attended Glasgow Dental Hospital with an acute painful swelling in the lower left jaw. She had previously (4 weeks prior) been treated with 7 days penicillin and the day before she presented, she had been given achromycin by her medical practitioner.

On clinical examination, an acute dental abscess was diagnosed associated with retained $/\overline{5}$ root in the otherwise edentulous mouth.

The abscess was incised under general anaesthesia and the root removed. A 3 week course of achromycin was instituted on bacteriological diagnosis of actinomycosis. Resolution was quickly achieved.

Case 11

34 year old male Caucasian (chemist) This patient presented at Glasgow Dental Hospital with a large

swelling of the right face centred over the right temporomandibular joint. It had been present for 1 week and a severe trismus had developed progressively. Although clinical examination was of necessity limited intra-orally, a pericoronal infection of the partially erupted $\overline{8}7$ with submasseteric abscess formation was diagnosed. Radiographs confirmed the impaction of $\overline{8}7$ and revealed no other source of infection.

Penicillin was prescribed initially with little success, and erythromycin was substituted. Over the next 10 days the swelling reduced slightly, the trismus became less profound, and the patient became more comfortable. The antibiotic was withdrawn after a 10 day course and the swelling reduced progressively and was noted to descend towards the angle of the jaw.

Two weeks later, reddening was noted superficially, and kaolin poultices were administered in an attempt to encourage pointing of the abscess. Six days after the reddening first appeared, the abscess discharged spontaneously, and pus was collected. Further surgical investigation was carried out under general anaesthesia the same day to ensure total evacuation of the abscess.

Bacteriological examination revealed the infection to be actinomycosis, and a total of 5 weeks penicillin was prescribed. Good resolution was achieved over this period and on the sixth week, the impacted $\overline{8}7$ was surgically removed under local anaesthesia. Erythromyicin was used as a prophylactic anti-microbic over a 10 day period. Healing proved uneventful.

Case 12

22 year old female Caucasian (student) The patient was referred for surgical opinion having been treated for a pain dysfunction syndrome of the right temporomandibular joint. When examined, a tense painful swelling in the region of the right cheek and parotid area was noted. Pus could be expressed through the orifice of the right parotid duct, and there was a severe trismus which rendered intra-oral examination difficult. Penicillin therapy was instituted and two weeks later the abscess was incised and drained extra-orally. Bacteriological examination of the pus revealed a mixed infection heavy in Eikenella corrodens but with A. israelii also present. Fluorescent antibody testing of the 3 day anaerobic culture confirmed this latter organism's presence. Clear saliva was noted 24 hours after incision.

The patient was reviewed regularly over the succeeding months, during which penicillin V, erythromycin, clindamycin and cephalexin were used for periods of 6, 5, 2.5 and 6 weeks respectively. Discharge from the wound continued for some 10 weeks but became more scant progressively. The trismus was steadily but slowly relieved. The resolved abscess caused some scarring due to severe fibrosis but the patient was unwilling at that time to consider plastic surgery. Sialograms taken 11 weeks after cessation of antibiotic therapy showed only minor areas of gland damage.

Case 13

34 year old Caucasian male (sales manager) The patient was referred to the Glasgow Dental Hospital by a general hospital with a large painful swelling in the right cheek. He had first noticed pain in this area some 10 weeks previously, but the swelling had only developed within the last 3 weeks. He had reported ulceration of the cheek in this area when a new partial upper denture had been fitted before any discomfort had started. No dental focus was found.

Treatment involved incision and drainage under general anaesthesia with liberation of copious pus. Immediate bacteriological examination was carried out, and a positive fluorescent antibody test confirmed actinomyces israelii. The patient was given intra-muscular penicillin and was maintained on oral penicillin for 5 weeks. Resolution was rapid and the trismus was relieved markedly within 2 days.

Case 14

The patient was a 13 year old Caucasian schoolgirl who was referred by her dental practitioner with an acute left submandibular abscess related to a carious /6. The appearance of the swelling allowed a provisional diagnosis of cervico-facial actinomycosis to be made. The swelling was of 2-3 weeks in duration, and had been unsuccessfully treated with penicillin.

Under general anaesthesia, the /6 was extracted and the abscess was incised. Immediate bacteriological

testing with Gram and fluorescent stain confirmed the clinical diagnosis, and the patient was treated with oral penicillin for 3 weeks, followed by erythromycin for a further 2 weeks. Resolution was achieved, though some scarring remained on the skin surface. This was considered due to the florrid complexion of the child.

Case 15

A 32 year old Caucasian sales representative was referred from the Periodontology Department of Glasgow Dental Hospital and School with an acute alveolar abscess associated with /$\overline{5}$. This tooth was extracted uneventfully, but 10 days later he returned with severe pain in this region. The /$\overline{6}$ was suspected to have a pulpitis, and a dressing was therefore inserted in this tooth, the /$\overline{5}$ socket was dressed with a small Whitehead's Varnish (Co. Pig. Iodoform) pack, and oral penicillin V was prescribed.

He was seen periodically over the ensuing 3 weeks, and during this time the buccal sulcus /$\overline{56}$ area became progressively more swollen, until fluctuation was elicited and the abscess was incised intra-orally. Culture of this discharge revealed a mixed infection of streptococcus viridans and haemophilus influenzae. Erythromycin was prescribed and improvement was noted within 24 hours. Some 2½ weeks later, however, the swelling recurred, but mainly related to the body of the mandible in the /$\overline{56}$ region. The /$\overline{6}$ was extracted, and ampicillin prescribed together with instructions to

use saline mouthwashes copiously. Radiographs at this time showed small punched out areas of rarefaction in the body of the mandible in the $\overline{5-8}$ region. A diagnosis of subperiosteal osteomyelitis was made, and immediate arrangements were instituted to remove the infected bone surgically.

The operation was carried out under intubation general anaesthesia, and ceporex was prescribed over the following week.

Healing post-operatively was good, but a hard soft tissue nodule was palpated in the left submandibular region which persisted and moved towards the mid-line over a period of 3 months. The swelling was painless, and failed to respond to further antibiotic courses of ceporex and tetracycline. After this 3 months, however, radiographs confirmed the clinical impression that the bone had healed. A further 3 months elapsed during which time the swelling established itself in the submental region but remained static. It was finally decided to re-admit the patient for surgical dissection of this nodule but before this could be done, the swelling presented on the skin surface, and discharged through multiple sinuses. A pus sample was taken, and a diagnosis of actinomycosis was made on bacteriological examination of the granules present.

Penicillin therapy was instituted immediately, and the drainage was encouraged with magnesium sulphate dressings. Penicillin was prescribed for 6 weeks.

Appendix B

Precis of Case Reports of Intra-oral Actinomycosis

Case A

A 39 year old female Caucasian was referred by her dental practitioner with pain in the lower anterior segment. She had undergone removal of the lower anterior teeth 11 weeks previously. The pain was constant and was exacerbated by palpation in the labial sulcus. A small sinus was noted in the $\overline{2/}$ region and radiographs of the $\overline{3-/-3}$ region revealed an ill defined area of radiolucency consistent with the provisional clinical diagnosis of osteomyelitis. The patient was given 500 mg. lincomycin intra-muscularly and 2 days later this was repeated. The acute symptoms subsided and a curettage was undertaken under local anaesthesia, 4 days after her initial presentation. At this time, the provisional clinical diagnosis was amended to an infected residual dental cyst.

Histopathological examination of the lesion revealed it to be a keratocyst with numerous bacteria within the lumen, predominantly colonies of Gram-positive branching filaments consistent with actinomyces. A diagnosis of kerato cyst was a superimposed actinomycotic infection was hence established. The patient was seen at intervals over the following 8 months during which time healing proved uneventful and new dentures were satisfactorily provided. Further progress could not be established due to the patient breaking repeated appointments.

Case B

The patient was a 40 year old female Caucasian.

She attended the Periodontology Department of the Glasgow Dental Hospital with an "inflamed gum" in the /34 region. A swab from the interdental region was reported by the microbiology department as containing a Gram positive filament which could be an actinomyces, although culture was not possible. After 9 weeks conservative treatment, a gingivectomy was carried out but this failed to effect resolution, and the patient returned after 6 months, when the /45 were extracted due to bone loss in the region related to trauma in lateral mandibular excursion. Pathological examination of the tissue revealed actinomyces like micro-organisms in the attached granulation tissue.

The patient was recalled in 4 months when healing was seen to be satisfactory.

Case C

A 6 year old Caucasian schoolgirl presented complaining of pain in the upper right quadrant of the mouth of 3 days duration. On examination a partially erupted 5/ was noted which discharged pus on digital pressure. Radiographic examination revealed a carious incompletely formed tooth and this was extracted under general anaesthesia on the day of presentation.

Histopathological examination of the decalcified tooth revealed micro-organisms consistent with actinomyces within the pulp.

Case D

A 21 year old female Caucasian presented complaining

of a persistent palatally discharging sinus in the 6/ area. The tooth had been root treated some 4 years previously and radiographically there was a distinct area of periapical radiolucency consistent with an apical dental cyst. Accordingly, it was decided to extract this tooth under local anaesthesia followed by a curettage of the infected cyst cavity.

The curettings were examined histopathologically and colonies of micro-organisms were noted which were consistent with actinomyces. A swab was taken of the wound 2 weeks later and actinomyces naeslundii was isolated bacteriologically. Healing, however, was unimpeded and a check radiograph 3 months later showed normal bone regeneration.

Case E

The patient was a 65 year old female Caucasian. This lady had undergone surgery in the right mandible for removal of an ossifying fibroma 5 years previously. This had recurred, and the lesion was removed under intubation general anaesthesia.

Pathological examination of the excised tissue revealed the lesion to be Paget's disease with organisms morphologically and tinctatorially consistent with actinomyces. The wound failed to heal, and was re-curetted surgically 48 days later. Once again, actinomyces were reported from the paraffin sections.

Case F

This patient, a 76 year old male Caucasian, underwent

removal of a retained root. The patient had previously been diagnosed as having Paget's disease which affected the jaws. The root was examined along with associated granulation tissue and a portion of sequestrum. The pathologist reported actinomyces-like micro-organisms related to the bone fragment which showed the typical histological appearance of Paget's disease.

Case G

Female (Caucasian), 18 years of age. The patient presented with a complaint of pain and swelling in the lower right jaw. Examination clinically revealed a bony swelling buccal of the alveolus in the $\overline{65}$/ region. A sinus was present through which pus could be expressed on pressure. A swab was taken of this discharge and the patient was radiographed. The x-rays revealed an ill defined patchy radiopacity contrasting with radiolucency and consistent with the appearance of a complex composite odontome in the $\overline{65}$/ area. An incidental finding was an unerupted $\overline{5}$/ lying in horizontal position with the crown anteriorly. Bacteriological examination of the pus revealed an actinomyces species in a mixed bacterial flora.

The odontome was removed surgically under penicillin cover, and the antibiotic was maintained during the post-operative phase. Healing was good, and the general medical practitioner agreed to continue penicillin therapy for 6 weeks. Histopathological examination of the excised lesion confirmed the clinical diagnosis of a complex

odontome, and reported actinomyces-like organisms associated with a mixture of acute and chronic inflammatory cells within the odontome.

Follow-up was not possible due to the patient not keeping appointments.

Case H

The patient, a 41 year old male Caucasian, presented with a hard swelling in the lower labial sulcus associated with $\overline{21/12}$. Radiographs revealed an apical dental cyst involving the apices of these four incisors, and the teeth were extracted together with enucleation of the cyst. Both $\overline{1/1}$ were non-vital.

The pathology report indicated that masses of Gram positive branching filaments were present within the cystic cavity, and were consistent with actinomyces. No further clinical details were obtained.

Case J

A 25 year old female Caucasian attended the Glasgow Dental School complaining of a recurrent swelling in the left cheek. This had arisen on 3 occasions since extractions had been performed in the left mandibular molar quadrant 1 year previously. The most recent episode had started 2 weeks prior to her presentation and oral penicillin which had been prescribed by her medical practitioner had failed to effect resolution. Clinical examination revealed a small round nodular swelling (approximately 1 cm. in diameter) within the substance of the cheek just anterior to the masseter insertion

to the lower border of the mandible, whilst radiographs showed retained /67 roots. A diagnosis of chronic adenitis of a facial lymph node was made and the following day the roots were removed and the "lymph node" dissected out under local anaesthesia. Histopathological examination of the lesion revealed an actinomycotic abscess with striking actinomycotic granules within the inflammatory exudate. The patient returned 6 days later for removal of sutures and a 7 day course of terramycin was prescribed. Resolution was slow and some 5 weeks after surgery a further 2 week course of terramycin was given. Resolution was uneventful therafter and the patient was discharged.

Case K

A 70 year old female Caucasian attended the Dental Hospital complaining of a swelling in the upper right jaw. On examination, a sinus was noted in the 4/ region, and radiographs revealed a residual dental cyst. After a short course of penicillin V, the cyst was surgically enucleated, and the pathology report indicated actinomyces-like filaments within the cyst. Healing was unimpeded, and 4 weeks later the patient was discharged.

Case L

A 35 year old Caucasian female presented with a swelling in the floor of the mouth of 4 months duration. It was not painful and had altered little in size since it had first been noticed. On examination, a small round

firm swelling approximately 0.5 cm. in diameter was noted to the right side of the lingual fraenum. A provisional diangosis of a mucous extravasation cyst which had undergone fibrosis was made.

The swelling was excised under local anaesthesia and the histopathological examination reported the lesion as a lymphoepithelial cyst containing bacteria morphologically consistent with actinomyces species.

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