NOVEL ASPECTS OF COMMONLY ENCOUNTERED PITUITARY ADENOMAS IN CLINICAL PRACTICE

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Abstract

Clinically relevant pituitary adenomas are common in the general population occurring in 1 per 865-1470 people. Prolactinomas and non-functioning pituitary adenomas account for more than 80% of pituitary adenoma types. My thesis is composed of several studies, undertaken at a single centre specialising in the multidisciplinary care of pituitary diseases, investigating clinical issues pertinent to the management of patients with the above adenomas. The findings of these studies contribute to the understanding of outcomes in the local practice, as well as being an Australian contribution to the international experience of managing these adenomas.

Firstly, I examine the issue of valvular heart disease in prolactinoma patients treated with cabergoline therapy. By studying a local cohort and by a systematic review I found that the prevalence of cabergoline-associated valvular heart disease (CAV) in prolactinoma patients to be extremely low, with just three confirmed cases amongst 1800 patients described in the literature. I have shown evidence that a simple annual cardiovascular examination is a suitable screening tool for this rare condition and made recommendations on when to consider a diagnostic echocardiogram.

Secondly, I examine two clinical issues affecting patients with surgically treated non-functioning pituitary macroadenomas (NFPMAs): the issues of regrowth and recurrence, and of hormonal outcomes. In the largest Australian cohort of cases with NFPMAs to be described, it was found that residual disease is a common finding post-surgery. In cases with residual disease, regrowth occurred in 40% of these cases compared to only 12.5% in cases without residual disease, at a median of 3.6 years of follow-up. Not surprisingly, larger baseline adenoma size was a predictor of regrowth and recurrence. In multivariate analysis, the presence of residual disease and younger age (under 41 years) at presentation were independent risk factors for regrowth and
recurrences. Based on this risk factor of younger age, I have recommended that these cases have lifelong radiological follow-up.

Results of hormonal outcomes in cases with NFPMAs treated with surgery showed the novel finding of gender differences in hormonal outcomes, with males having a higher prevalence of multiple (≥2) hormone deficiencies (MHD) at presentation than females, and after surgery. In particular, pre-menopausal females had very few hormone deficiencies post-operatively and demonstrated a propensity to recover function. These gender subgroup differences are the first to be noted in the literature and have important clinical implications particularly for pre-menopausal females where fertility preservation is an issue. I also found that pre-menopausal females had smaller adenomas than males suggesting that they may present earlier in the natural history of the disease. In multivariate analysis, larger adenoma size remained a significant factor associated with post-operative MHD.

The final part of this thesis is an exploratory study of intrasellar pressure (ISP) and how it relates to adenoma size and hormonal outcomes in cases with NFPMAs undergoing surgery. I did not find that ISP was correlated to adenoma size, but results did suggest that raised ISP may play a role in more frequent hormone deficiencies at presentation. In this study adenoma size was again noted to be showing a trend towards influencing hormonal outcomes. The findings from this study will require a larger cohort in order to better quantify how ISP and adenoma size interact to contribute to the pathophysiology of hormone deficiencies.

In these three studies of NFPMAs adenoma size has emerged as an important factor influencing surgical and hormonal outcomes; smaller adenomas are more likely to be fully resected and less likely to result in hormone deficiencies. This thesis lays the foundations for advocating for the consideration of early surgical intervention of NFPMAs (when adenomas are smaller and earlier in the natural history of the disease),
in order to optimise surgical and hormone outcomes. The above should now be an important clinical consideration in the multidisciplinary care of these patients.
Declarations

This is to certify that

(i) The thesis comprises only of original work except where indicated in the preface,

(ii) Due acknowledgement has been made in the text to all other material used,

(iii) The thesis is fewer than 100,000 words in length, exclusive of tables, maps, bibliographies and appendices

Name: Carmela Caputo

Signature:

Date: 12 December 2019
Preface

This research was carried out under the supervision and guidance of Professor Richard MacIsaac, Associate Professor Warrick Inder and Mr Yi Yuen Wang at St Vincent’s Hospital Melbourne.

The structure of this thesis is as follows...

1) The literature review (Chapter 1);
2) Chapters which comprise of original papers published in peer-reviewed journals (Chapter 2, 3 and 4);
3) A further chapter containing unpublished material not submitted for publication (Chapter 5);
4) Summary of the thesis findings (Chapter 6);
5) Conclusions and future directions (Chapter 7);
6) References;
7) Appendices.

I was responsible for...

1) Development of all project conception and methods;
2) Preparing and submitting ethics approval for all projects;
3) The execution of each study;
4) Collection of data for all studies;
5) Recruitment of participants for the prospective studies;
6) Analysis of data for each study;
7) Preparation of published manuscripts.
The following people have assisted in my research:

Chapter 2: Professor David Prior, The Cardiology Department, St Vincent’s Hospital, Melbourne, for overseeing the echocardiograms performed in this study by his department.

Chapter 3: Dr Anna Watts, for the additional collection of data from 2010-2013 to add to the pre-existing data from 1995-2010 collected by myself. Together with Dr Watts, we contributed to data analysis and to the writing of manuscripts towards final publication. Statistical advice was contributed by Mr Abhishek Easwaren, Department of Biostatistics, St Vincent’s Hospital Melbourne.

Chapter 4: Statistical advice on multivariate analyses was contributed by Professor Stephen Farish, The University of Melbourne.

Chapter 5: Mr Yi Yuen Wang and Mr Peter McNeill for the active recruitment of participants and for performing the intrasellar pressure in the pituitary adenomas. Statistical advice was contributed by Dr Graham Hepworth, The University of Melbourne, and Associate Professor Esther Briganti.

Five manuscripts have been published and are included in this thesis as Chapter 2 Publications 1, 2, and 3, Chapter 3 and Chapter 4. All co-authors were involved in the revision of manuscripts.
This thesis requires acknowledgement from the following...

1) Australian Government Research Training Program Scholarship for subsiding this higher research degree;
2) St Vincent’s Hospital Research Endowment Fund;
3) The Pegasus Foundation;
4) The Endocrine Society of Australia.

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- **St Vincent’s Hospital Research Endowment Fund**: funding for the intrasellar pressure probes. This funding enabled me to start the study for Chapter 5.

- **Pegasus Foundation and Mr Yi Yuen Wang**: funding for the intrasellar pressure probes. This funding enabled me to complete the study for Chapter 5.

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I would like to thank all the participants of the various studies, without whom, these studies would not exist.
I would like to thank my family and friends (Con, Stephanie, Mauro, Mary and my parents) for all your support and patience over this time. Above all, thank you God, the Divine Creator whom gave us His only begotten Son.
Third party copyright material

No third-party copyright material has been included in the thesis.
Chapter 1: The literature review

1.1 The pituitary gland

The pituitary gland secretes several vital hormones required for life processes and functions. It is situated in the middle cranial fossa and in the sella turcica (or pituitary fossa). The gland measures on average 4.8mm in maximum height (range of 1.6-8.6mm) and is usually taller in adolescence compared to those above 20 years of age, and higher in females compared to males (1) (2). The mean volume of the normal pituitary gland has been measured between 602mm$^3$ (3) and 639mm$^3$ (2) in females, and with a mean of 495mm$^3$ in males (2). In pregnancy the pituitary gland enlarges due to hypertrophy of lactotroph cells to enable lactation, with maximum heights being reached in the in the first week post-partum (mean 9.3mm, range 6.5-11.8), but then rapidly decreasing back down to the pre-pregnancy size (4).

The pituitary gland consists of two parts, the anterior and posterior pituitary gland which are derived embryologically from the oral ectoderm and neural ectoderm respectively, during the fourth week of development resulting in distinctive cell types and function. The posterior pituitary gland being of neuronal origin consisting of nerve endings which originate in the hypothalamus (supraoptic and paraventricular nuclei) and release the hormones oxytocin and vasopressin which are required for parturition and water homeostasis, respectively. The anterior pituitary gland consists of differentiated cells which secrete hormones and structural supportive cells. The anterior pituitary gland secretes the following hormones, growth hormone (GH), adrenocorticotropic hormone (ACTH), the gonadotropins-follicular stimulating hormone (FSH) and luteinising hormone (LH), thyroid stimulating hormone (TSH) and prolactin. The release of these hormones results in critical end organ effects including growth and development, pubertal induction, secondary sexual characteristics, fertility, cardiovascular and immune functions.
The pituitary gland is connected to the brain via the pituitary stalk arising from the hypothalamus (Figure 1.0 A and B). The secretion of hormones from the anterior pituitary gland and the release of preformed hormones from the posterior pituitary gland, is highly regulated from the hypothalamus with direct feedback from the periphery.
Figure 1.0 A and B. These are the normal MRI findings of 23 year old female. The pituitary gland measures 9.7mm anterior-posterior, 5.4 mm height and 12.0mm width.

A: MRI coronal view, white arrow - pituitary gland, green arrow - pituitary stalk, blue arrow - the optic chiasm, red arrows - the carotid arteries in the cavernous sinuses.

B: MRI sagittal view, white arrow - the pituitary gland with stalk above, blue arrow - optic chiasm, yellow arrow - the hypothalamus.
1.2 Pathologies of the pituitary gland

The pituitary gland can be affected by several pathologies. These include pituitary tumours (of which pituitary adenomas are the commonest form), congenital abnormalities, infiltrative disease processes, apoplexy and ischaemia.

Congenital abnormalities are rare and varied in pathologies. They include conditions such as septo-optic dysplasia where there is malformation of facial and cranial midline structures causing a hypoplastic pituitary gland (5), the pituitary stalk interruption syndrome where there can be absence or a thin pituitary stalk associated with a hypoplastic and ectopic anterior and posterior pituitary gland, and tumour susceptibility syndromes that can be associated with pituitary tumours at a very young age such as the DICER1 syndrome. In many of these congenital conditions, the causative genetic abnormalities are being increasingly discovered.

Infiltrative pituitary diseases can occur due to a number of different pathologies. Hypophysitis is one form of infiltrative process where there is enlargement and inflammation of the pituitary gland and/ or pituitary stalk. Hypophysitis is rare with an annual incidence of 1 in 7-9 million and accounts for <1% of pituitary surgeries (6). Hypophysitis can be due to primary or secondary causes. Primary causes include lymphocytic hypophysitis, which has a female preponderance, and secondary causes due other systemic diseases such as Langerhans’s histiocytosis (Figure 1.1 A and B), IgG-4 disease (Figure 1.2 A and B), sarcoidosis and Wegener’s granulomatosis (6).
Figure 1.1 A and B. Langerhans’s histiocytosis. A 32 year old female was diagnosed with panhypopituitarism. Her MRI demonstrates a homogeneously enhancing suprasellar mass involving the hypothalamus and pituitary stalk, measuring 16 x 8 x 16mm (blue arrows). The mass abuts the optic chiasm. Biopsy of the suprasellar lesion demonstrated features consistent with Langerhans’s histiocytosis.

A: MRI coronal view

B: MRI sagittal view
Figure 1.2 A and B. IgG-4 disease of the pituitary gland and stalk in a 49 year old male. The patient presented with panhypopituitarism and diabetes insipidus. Post-contrast MRI of the pituitary demonstrating homogenously enhancing non-tapering pituitary stalk and an expanded, enhancing pituitary with loss of the normal posterior pituitary ‘bright spot’ (7).

A: MRI coronal view, blue arrow - expanded pituitary gland and stalk

B: MRI sagittal view, blue arrow - expanded pituitary gland and stalk with involvement of the disease into the floor of the third ventricle
A recent secondary cause of hypophysitis has been described caused by the immune checkpoint inhibitors (8). These new classes of drugs are used for cancer therapy and work by stimulating the immune response to activate T Cells which leads to a cascade of events to attack cancer cells (especially melanoma and renal cell cancers). Side effects of these treatments are autoimmune endocrine diseases including hypophysitis, thyroiditis and adrenalitis. Hypophysitis occurs in 8-11.7% of those taking these drugs (8). Presentation of immune checkpoint hypophysitis is similar to other forms of hypophysitis with symptoms related to hormone deficiencies, headaches or mass effect.

Other forms of infiltrative processes include infection (such as tuberculosis) and iron deposition (such as hemochromatosis). Metastatic deposits to the pituitary gland, most frequently breast and lung cancer, account for approximately 1% of pituitary surgeries (9).

Pituitary apoplexy is a clinical syndrome that occurs as a result of acute haemorrhage and/or infarction into a pituitary adenoma. It is an uncommon event occurring in 0.6-9.1% of neurosurgical series of pituitary adenomas undergoing surgery (10). The sudden volume expansion of the pituitary adenomas at the time of apoplexy results in the classical presentation of sudden onset headache which can be associated with photophobia, cranial nerve palsies and multiple hormone deficiencies (Figure 1.3 A and B).
Figure 1.3 A and B. Pre-operative MRI findings of a 28 year old male presenting with headache and right third cranial nerve palsy due to apoplexy. The adenoma measures 27 x 24 x 19mm with elevation and compression of the optic chiasm. The MRI appearances with the heterogeneous changes are in keeping with central degeneration and hyperintensity due to blood products. This case underwent urgent surgery with optimal outcomes of complete tumour resection, reversal of visual and cranial nerve abnormalities and full preservation of pituitary hormone function.

A: MRI coronal view, blue arrow - apoplexy

B: MRI sagittal view, blue arrow - apoplexy
Sheehan’s syndrome is another condition caused by infarction of the pituitary gland occurring after a post-partum haemorrhage. This event is associated with a high prevalence of acute and latent pituitary hormone deficiencies (3). If unrecognised some cases can prove fatal secondary to untreated cortisol deficiency. Due to improved obstetric care in the developed world Sheehan’s syndrome is an uncommon occurrence these days (11). Another unusual cause of ischaemia to the pituitary gland is when its vascular supply is compromised by carotid artery aneurysms and its treatment by coiling (12). An example of this is shown in Figure 1.4.

*Figure 1.4* The MRI findings of a 50 year old female who required stenting of bilateral cavernous sinus aneurysms. This inadvertently resulted in ischaemia of the pituitary gland resulting in the patient developing pituitary hormone deficiencies (12). Straight white arrow depicts the right coil in the cavernous sinus, and the curved arrow (left), depicts pituitary gland displaced to the left.
Despite the varied aetiologies that can affect the pituitary gland, as described above, a common finding in all these conditions is the association with impaired hormone function leading to one or more pituitary hormone deficiencies.
1.3 Pituitary tumours

Tumours arising from the pituitary gland are derived from functioning or supporting cells within the pituitary gland or rarely from cells that have migrated from elsewhere in the body. The 4th edition of the World Health Organisation (WHO) classification of pituitary tumours classifies these tumours as listed in the table below (13) (Table 1.0).

Pituitary adenomas are the third commonest intracranial tumour after meningiomas and gliomas and account for 10-15% of intracranial tumours (14). Pituitary carcinomas are defined by the presence of distant metastases; these are rare, accounting for 0.1% of pituitary adenomas. Outcomes in those with pituitary carcinomas are universally poor (15).

The figures below are examples of two pituitary tumours encountered in my clinical practice. Figure 1.5 A and B show the MRI findings of a case with an adamantinomatous craniopharyngioma and Figure 1.6 A and B show the MRI findings of a case with a mixed gangliocytoma-adenoma (16).

The remainder of this literature review will focus on pituitary adenomas summarising the different types, their epidemiology and management.
Table 1.0 2017 WHO classification of pituitary tumours (13)

<table>
<thead>
<tr>
<th>Classification of pituitary tumours</th>
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<tbody>
<tr>
<td><strong>Pituitary adenomas</strong></td>
</tr>
<tr>
<td>Somatotroph adenoma</td>
</tr>
<tr>
<td>Lactotroph adenoma</td>
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<tr>
<td>Thyrotroph adenoma</td>
</tr>
<tr>
<td>Corticotroph adenoma</td>
</tr>
<tr>
<td>Gonadotroph adenoma</td>
</tr>
<tr>
<td>Null-cell adenoma</td>
</tr>
<tr>
<td>Plurihormonal and double adenomas</td>
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<tr>
<td><strong>Pituitary carcinoma</strong></td>
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<tr>
<td><strong>Pituitary blastoma</strong></td>
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<tr>
<td><strong>Tumours of the posterior pituitary</strong></td>
</tr>
<tr>
<td>Pituicytoma</td>
</tr>
<tr>
<td>Granular cell tumour of the sella</td>
</tr>
<tr>
<td>Spindle cell oncocytoma</td>
</tr>
<tr>
<td>Sellar ependymoma</td>
</tr>
<tr>
<td><strong>Neuronal and paraneuronal tumours</strong></td>
</tr>
<tr>
<td>Gangliocytoma and mixed gangliocytoma-adenoma</td>
</tr>
<tr>
<td>Neurocytoma</td>
</tr>
<tr>
<td>Paraganglioma</td>
</tr>
<tr>
<td>Neuroblastoma</td>
</tr>
<tr>
<td><strong>Craniopharyngioma</strong></td>
</tr>
<tr>
<td>Adamantinomatous craniopharyngioma</td>
</tr>
<tr>
<td>Papillary craniopharyngioma</td>
</tr>
<tr>
<td><strong>Mesenchymal and stromal tumours</strong></td>
</tr>
<tr>
<td>Meningioma</td>
</tr>
<tr>
<td>Schwannoma</td>
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<tr>
<td>Chorda, NOS</td>
</tr>
<tr>
<td>- Chondroid chordoma</td>
</tr>
<tr>
<td>- “De-differentiated” chordoma</td>
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<tr>
<td>Solitary fibrous tumour/ hemangiopericytoma</td>
</tr>
<tr>
<td>Grade 1 SFT/HPC</td>
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<tr>
<td>Grade 2 SFT/HPC</td>
</tr>
<tr>
<td>Grade 3 SFT/HPC</td>
</tr>
<tr>
<td><strong>Hematolymphoid tumours</strong></td>
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<tr>
<td><strong>Germ cell tumours</strong></td>
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<td>Germinoma</td>
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<tr>
<td>Yolk sac tumour</td>
</tr>
<tr>
<td>Embryonal carcinoma</td>
</tr>
<tr>
<td>Choriocarcinoma</td>
</tr>
<tr>
<td>Teratoma, NOS</td>
</tr>
<tr>
<td>Mature teratoma</td>
</tr>
<tr>
<td>Immature teratoma</td>
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<tr>
<td>Teratoma with malignant transformation</td>
</tr>
<tr>
<td>Mixed germ cell tumour</td>
</tr>
<tr>
<td><strong>Secondary tumours</strong></td>
</tr>
</tbody>
</table>

25
Figure 1.5 A and B. A craniopharyngioma measuring 38 x 45 x 39mm in a 16 year old female patient. Although these pituitary tumours are benign, they are often extremely large and cause major problems to vision and hormone function of the pituitary gland and hypothalamus. This patient has had successful pituitary surgery via the transsphenoidal route and remains in remission at 7 years follow-up. The patient however is legally blind and has panhypopituitarism with diabetes insipidus.

A: MRI coronal view, blue arrow - giant craniopharyngioma

B: MRI sagittal view, blue arrow - giant craniopharyngioma
Figure 1.6 A and B. A 60 year old male presented with headaches and was found to have biochemical evidence of excessive growth hormone. The MRI shows a 19 x 17 x 24mm lesion in the right side of the pituitary gland. After surgery histology demonstrated a case of a neuronal tumour of mixed gangliocytoma with a pituitary adenoma secreting growth hormone (16).

A: MRI coronal view, blue arrow - mixed gangliocytoma

A: MRI sagittal view, blue arrow - mixed gangliocytoma
1.4 Pituitary adenomas

Pituitary adenomas occur through clonal proliferation of a single cell line (17). In autopsies and radiologic imaging studies in the general population pituitary adenomas are relatively common findings, with a study reporting a prevalence of 17% in the general population (18) (19): although clinically relevant adenomas occur in 0.1% of the population (20).

In approximately 60% of cases of pituitary adenomas the cause is unknown. However, with increasing use of genetic techniques, mutations are being discovered at an impressive rate. In approximately 40% of pituitary adenomas, somatic mutations have been described, including GNAS mutations in growth hormone producing adenomas (in somatotropinomas), and more recently USP8 mutation in corticotropic adenomas (21). In approximately 5% of pituitary adenomas are due to well described germline mutations (22). These germline mutations are currently identified in the following syndromic conditions:

1) Familial Isolated Pituitary Adenoma (FIPA)
2) Multiple Endocrine Neoplasia Type 1 (MEN-1)
3) Multiple Endocrine Neoplasia Type 4 (MEN-4)
4) Carney Complex
5) “3P” syndromes of the succinate dehydrogenase deficiencies
6) Neurofibromatosis Type 1 (NF-1)
7) DICER1 syndrome

Less than 1% of pituitary adenomas are due to mosaic mutations resulting in McCune Albright disease and X-linked acrogigantism (XLAG) (21).
Pituitary adenomas are problematic as the following complications can occur: the production of hormone excess and complications related to this, hormone deficiencies, symptoms related to mass effect of the adenoma and long-term issues related to persistent or recurrent disease. Finally, although not common, the potential of the adenoma to behave aggressively or develop into a carcinoma.
1.5 Pituitary adenoma types

The classification of pituitary adenomas is changing with time due to the increased understanding of the pathophysiology, genetic causes being identified and with improved histological techniques (23).

Traditionally, pituitary adenomas have been characterised by function (clinical manifestation and biochemical evidence of hormone excess) and with the support of histological diagnosis by immunohistochemistry (where histology has been obtained) (24). This traditional classification is still a useful broad classification used in the clinical office for face to face patient care (Table 1.1). Broadly, these adenomas can also be classified as hormonally functioning (due to an excess of pituitary hormone produced), and hormonally inactive (non-functioning pituitary adenomas).

A description of these pituitary adenoma types will follow, with emphasis on prolactinomas and non-functioning pituitary adenomas, as these are the adenoma types which are the focus of this thesis.

Table 1.1 Traditional classification of pituitary adenomas

<table>
<thead>
<tr>
<th>Adenoma type</th>
<th>Hormone produced</th>
<th>Name of clinical disease</th>
</tr>
</thead>
<tbody>
<tr>
<td>Prolactinoma</td>
<td>Prolactin (PRL)</td>
<td></td>
</tr>
<tr>
<td>Non-functioning pituitary adenoma</td>
<td>Nil</td>
<td></td>
</tr>
<tr>
<td>Somatotropinoma</td>
<td>Growth hormone (GH)</td>
<td>Acromegaly</td>
</tr>
<tr>
<td>Corticotropinoma</td>
<td>Adrenocorticotrophic hormone (ACTH)</td>
<td>Cushing’s Disease</td>
</tr>
<tr>
<td>Thyrotropinoma</td>
<td>Thyroid Stimulating Hormone</td>
<td></td>
</tr>
<tr>
<td>Gonadotropinoma</td>
<td>Follicular (FSH) or luteinising (LH) hormones</td>
<td></td>
</tr>
</tbody>
</table>
1.5.1 Prolactinomas

Prolactinomas are the commonest type of pituitary adenomas both in incidence and prevalence. Prolactinomas secrete excessive amounts of prolactin. Prolactinomas are categorised by size, with those <10mm classified as microprolactinomas (Figure 1.6), those ≥10mm classified as macroprolactinomas and ≥40mm classified as giant prolactinomas. The diagnosis is made by elevated serum levels of prolactin which generally correlate with adenoma size. Prolactin levels associated with microprolactinomas are usually 2-5 times the upper limit of the normal value (with a normal prolactin level generally reported below 500mIU/L) whereas macroadenomas are associated with prolactin levels exceeding 5000mIU/L (25).

Prolactin is the one pituitary hormone which is predominantly under inhibitory control from dopamine signalling from the hypothalamus, contrary to the other pituitary hormones which are under stimulatory control. Physiological increases in prolactin occur in pregnancy due to the stimulatory effect of estrogen (26), physical or psychological stress and breast stimulation (27). There are many causes of mild hyperprolactinaemia other than prolactinomas, such as drug-associated (especially dopamine antagonists such as metoclopramide and antipsychotic agents), idiopathic, renal failure and disruption of the pituitary stalk by other pituitary lesions such as a non-functioning adenomas (28). This later effect is termed the “stalk effect”, and it has been shown that the maximum limits of prolactin in almost all cases with stalk effect do not exceed 2000mIU/L (29). Primary hypothyroidism can be associated with hyperprolactinemia due to the compensatory rise of thyrotropic stimulating hormone (TRH) from the hypothalamus having a stimulating effect on lactotrophic cells (30).
Figure 1.6 A 27 year old female presented with secondary amenorrhea and was found to have a raised serum prolactin level of 1400mIU/L (reference range 110-560). MRI post-contrast coronal view shows a right sided microprolactinoma 6 x 6 x 8mm (blue arrow).

In females (specifically pre-menopausal females), elevated prolactin levels lead to galactorrhoea, secondary amenorrhea and infertility. Due to the menstrual disturbances in pre-menopausal females, this gender subgroup is more likely to present earlier in the natural history of the disease compared to males (27) (31). In males with prolactinomas hyperprolactinaemia also results in hypogonadism and infertility, but males often present to medical attention due to mass effect (headaches and visual abnormalities) due to a higher prevalence of having macroadenomas.

Primary treatment of prolactinomas is with the oral medication called dopamine agonists (DAs). This class of medications work by stimulating dopamine receptor (specifically subclass receptor D2) on prolactin producing cells (lactotrophs) in the pituitary. This in turn, inhibits prolactin production and release from the lactotrophic cells and has anti-proliferative effects on these same cells. These drugs are highly
Efficacious at reducing adenoma size, prolactin levels and restoring gonadal function (28). A more detailed discussion of DAs will follow on page 35.

Surgery still has a role in the treatment of prolactinomas and historically was the treatment of choice until DAs were introduced in the late 1970s. The recognised indications for surgery are for prolactinomas that are resistant to treatment with DAs (defined as failure to normalise prolactin and/or reduction in adenoma size of large adenomas), cystic prolactinomas causing mass effect, adverse effects from medication, apoplexy and patient preference (32) (Figure 1.7).

Radiotherapy is another treatment modality that can also be used for hormonal reduction and mass control. Radiotherapy is most effective in controlling adenoma growth but less favourable at normalising prolactin levels (33). Radiotherapy is not generally considered first line treatment of prolactinomas but can be considered in selected cases that have not responded to medical or surgical treatment and those with malignant prolactinomas (34). Adverse effects of radiotherapy are discussed on page 52 of this thesis.
Figure 1.7 A, B and C. A 55 year old female presented with visual abnormalities. Her prolactin level was raised at 28,000mIU/L (reference range 110-560) and her MRI showed a large, irregularly shaped prolactinoma. Cabergoline treatment rapidly normalised her prolactin levels however there was minimal decrease in the size of the adenoma after 12 months of treatment (A: coronal view and B: sagittal view). The patient went on to have surgery due to the adenoma elevating the right optic nerve. Post-operatively there was some residual disease which has not grown over time (C: coronal view, blue arrow). Adenoma histology confirmed a prolactinoma with some areas of strong and weak staining of prolactin by immunohistochemistry. These histological features may explain why the adenoma may not have responded fully to cabergoline. Her post-operative prolactin level remains within the reference range and she has serial MRI scans for monitoring of the residual adenoma.
Medical therapy for prolactinomas

DAs are the medical treatment of choice for prolactinomas. There are several types of DAs that are used in the treatment of these adenomas. Introduced in the late 1970’s, bromocriptine was the first DA used to treat prolactinomas. It can reduce prolactin levels and adenoma size in 70-80% of cases (35). It has a relatively short half-life so needs to be given twice per day and can be associated with adverse effects of nausea and hypotension as it can bind to other subtypes of dopamine receptors besides the D2 receptors on lactrophic cells. Bromocriptine has been largely superseded by cabergoline, which has a much higher binding affinity to D2 receptors on lactrophic cells and has a long half-life compared to than bromocriptine; cabergoline subsequently has several advantages over bromocriptine - it is better tolerated and more convenient to take due to its weekly dosing regimen (Figure 1.8). Cabergoline has been reported to reduce prolactin levels and decrease adenoma mass in around 73-83% of cases (36). Both bromocriptine and cabergoline are ergot-derived DAs. A third DA, also ergot-derived, pergolide, is highly efficacious at reducing prolactin and inducing adenoma shrinkage (37); however, in Australia, the clinical use of pergolide for the treatment of prolactinomas has been limited as this class of DAs only had the clinical indication for the treatment of Parkinson’s disease. Finally, quinagolide is another DA however, is not an ergot-derived DA like the others above. In small studies quinagolide has been shown to be as effective and well tolerated as cabergoline but this medication has not been as extensively studied as cabergoline or bromocriptine, and clinical experience with quinagolide is limited. (38). Of note, quinagolide has just been withdrawn from the Australian market for commercial reasons (April 2019).

Medical therapy with DAs is generally for a prolonged period over years with varying doses of medication; although the optimum time period of treatment has not been well defined (39). There have been a limited number of studies examining when DAs can be withdrawn. The largest of these studies examined 105 cases with microprolactinomas and 70 cases with macroadenomas treated with cabergoline who had archived normalisation of prolactin levels and with complete disappearance of the
adenoma or with a >50% reduction in adenoma size on MRI scanning (40). In the cohort with microprolactinomas, who were treated with cabergoline for a median period of 48 months, 70% of cases maintained a normal prolactin level after cessation of therapy at a median of 36 months of follow-up. In the cohort with macroprolactinomas, who were treated with cabergoline for a median period of 42 months, 64% of cases maintained a normal prolactin level after cessation of therapy at a median of 48 months of follow-up (40). The 2011 Society of Endocrinology clinical practice guidelines for the diagnosis and treatment of hyperprolactinaemia suggest a minimum treatment period of two years before considering a trial off treatment in those cases who have archived a normal prolactin level and who have no visible adenoma on MRI scanning (28). Cessation of DA therapy can also be considered in post-menopausal females with microadenomas as it has been observed that post-menopausal females are more likely to normalise prolactin levels after dopamine withdrawal compared to pre-menopausal females. This is possibly explained by the declining estrogen levels in the post-menopause having less stimulatory effect on the lactotrophs (41). DA withdrawal in post-menopausal-females, however, is less likely to result in normal prolactin levels in cases with macroprolactinomas (41).
Figure 1.8 A, B, C and D. A 32 year old male presented with headaches. He was investigated with MRI detecting a pituitary lesion invading into the both left and right cavernous sinuses; A: MRI coronal view and B: MRI sagittal view. Biochemical testing demonstrated a markedly raised prolactin level of 90,000mIU/L (reference range 110-560) and a low testosterone level confirming the diagnosis of a macroprolactinoma. The patient has had excellent response to cabergoline with normalisation of prolactin and testosterone levels however this has taken several years to achieve this and requiring higher than average doses of cabergoline (approximately 4mg weekly). The latest MRI findings demonstrating excellent minimal residual disease, C: MRI coronal view, D: MRI sagittal view.
Dopamine agonists and valvular heart disease

In 2007, concern was raised about ergot-derived DAs, cabergoline and pergolide, being associated with fibrotic valvular heart disease in Parkinson’s disease (42) (43). These findings were of clinical significance to endocrinologists as these medications (namely cabergoline) are the mainstay of treatment of prolactinomas worldwide (28) (44). These increased risk of valvular disease were supported by other studies of Parkinson’s disease leading to the voluntary withdrawal of pergolide in March 2007, whilst in July 2011 the United States food and drug administration (FDA) issued a new warning for cabergoline recommending regular echocardiograms every 6-12 months in all users of the drug.

Cabergoline-associated valvulopathy (CAV) represents a specific pathology characterised by the three salient echocardiographic features of moderate or severe valvular regurgitation, valve thickening and valve restriction (45). Furthermore, the absence of calcification and myxomatous changes distinguishes CAV from age-related sclerosis and myxomatous valve disease, respectively. If a valvular lesion is found it is crucial to distinguish between age-related changes, myxomatous change or CAV in order to determine if cabergoline should be ceased. The histological features of CAV (only available in those who have undergone surgical valve replacement) are characterised by a thickened valve due to fibrous proliferation with the absence of inflammatory cells, thrombus and calcification (46). The mechanism of valvular pathology as a cabergoline-induced adverse event is biologically plausible, via stimulation of serotonin (5-hydroxytryptamine [5-HT]) receptor subtype 5-HT(2B), which is expressed in heart valves with stimulation of these receptors being previously shown to mediate mitogenesis and proliferation of fibroblasts (47).

In a Parkinson’s cohort which developed valvular heart disease, the mean cumulative dose associated with this abnormality was 4015 mg (SD 3208); one standard deviation below this mean cumulative dose of cabergoline calculated at 720mg (43). These
cumulative doses of cabergoline in the Parkinson’s disease cohort with valvular heart disease is much higher than cumulative doses that occur in most patients treated for prolactinomas. Most prolactinoma patients are prescribed low dose cabergoline of 0.5-1.0mg per week and would reach a cumulative dose of 720mg after 26.7 years and 14.4 years, respective of each dose schedule. Only 10% of prolactinoma patients require high doses cabergoline treatment (≥3.0mg per week) (48); this later group would reach cumulative doses of 720mg of cabergoline after five years of treatment.

Since 2008, over 20 studies have been published reporting on the echocardiographic findings of prolactinoma patients taking cabergoline. These studies are a combination of case-control studies, cross-sectional studies and prospective follow-up studies. The details of these studies are presented in a systematic review in Chapter 1 (Publication 1 of my thesis). Despite all these studies, only one study has reported on the specific changes that are diagnostic of CAV on echocardiograms (49). The other studies have reported on non-specific valvular changes and therefore the true risk and prevalence of CAV in prolactinoma patients has been unclear. The original studies of valvular heart disease in the Parkinson’s cohort have also reported on non-specific valvular changes rather than the salient features of CAV (42) (43).

There have been three studies that have followed a total of 185 prolactinoma cases with serial echocardiograms after 2-5 years and have reassuringly not found an increased incidence of CAV (50) (51) (52). A recent community-based study from the UK demonstrated that very few cases with hyperprolactinaemia had baseline echocardiograms (2/45), and only 5/45 cases had serial echocardiograms two years apart (51). Even in Parkinson’s disease, a study has shown very few cases have had serial echocardiograms (53). Thus, echocardiography is seldom used, but more importantly, routine interval scanning has not been shown to be useful. More recently (2019), a meta-analysis based on 13 case control studies of approximately 800 prolactinomas patients concluded that cabergoline is associated with a risk of tricuspid regurgitation (54). The authors of this meta-analysis also suggested that more large-
scale, prospective, controlled, quality-assured echocardiographic studies were required and that additional cross-sectional studies drawn from routine practice were unlikely to inform the field significantly (54).

The clinical issue that arises from CAV in prolactinoma cases is how to best screen for this condition when echocardiograms have not been widely utilised in routine clinical practice and have a low yield for detecting CAV in follow-up studies. In 2011, the Endocrine Society guideline on the “diagnosis and treatment of hyperprolactinemia” recommended to consider echocardiography for those taking high doses of cabergoline and for prolonged periods (but exact dose and duration was not defined), and that echocardiograms would likely not be required for those at typically low doses of 1.0-2.0mg per week (28). These recommendations were based on the findings of seven case control studies of 500 prolactinoma patients at that time. This year (2019) the first dedicated guideline and recommendations for the use of echocardiograms for monitoring of patients taking cabergoline has been published jointly by the British Society of Echocardiography, the British Heart Valve Society and the Society for Endocrinology (45). These guidelines are based on the findings of the meta-analysis of 13 case control studies of approximately 800 prolactinoma patients mentioned in the previous paragraph (54), and recommended a baseline echocardiogram in all cases, followed by 5 yearly echocardiograms for those on low dose treatment (<2mg per week), but yearly echocardiograms for those taking ≥ 2mg per week (45).

In Chapter 2 of this thesis I will explore the clinical issue of CAV in prolactinoma patients by including the first Australian cohort to be studied. I will examine the prevalence of CAV by examining the literature and explore controversies about screening for this condition.
1.5.2 Non-functioning pituitary adenomas

Non-functioning pituitary adenomas are a heterogeneous group of adenomas that are characterised by the absence of secreted functional pituitary hormone. They present as incidental findings or symptomatically due to hormone deficiencies or mass effect of the adenoma (headaches or visual disturbance). These adenomas are heterogeneous as the immunohistochemistry varies with most staining for FSH and/or LH, others showing no positive staining (null cell adenoma) and those that stain for ACTH, GH, Prolactin or TSH, or a combination of these hormones.

Small pituitary non-functioning adenomas (microadenomas, <10mm) are commonly discovered incidentally on cerebral imaging for other reasons, whereas most large adenomas (non-functioning pituitary macroadenomas, ≥10mm, NFPMA's) are more likely to present with symptoms of mass effects or due to hormonal deficiencies (55). The 2011 Endocrine Society guidelines for the management of incidental pituitary adenoma recommend a systematic approach to management which includes clinical and biochemical assessment to detect pituitary hormone deficiencies or excess syndromes and assessment of visual pathways if the lesion is compressing the optic apparatus (56). Further imaging with MRI scanning with variable frequency depending on the size of the lesion: microadenomas being one year (with repeat hormone profiling), followed by 1-2 yearly thereafter, and six months for a macroadenoma (with repeat hormone profiling). These guidelines recommend surgery for those with adenomas which compromise to the optic apparatus, those with apoplexy or hormone excess. It is also recommended that surgery be considered for those that show significant growth, new hormonal loss, unremitting headache or pregnancy planning (56).

Observation is the usual management for small non-functioning pituitary adenomas, but surgery is often required for symptomatic patients with mass effect in NFPMA's (57) (58). A major management dilemma in the treatment of NFPMA's is that there is
high likelihood of having residual disease post-operatively and this is a risk factor for regrowth of the residual adenoma. Even if fully removed, NFPMAs can return (recurrence). Regrowth and recurrences may require further multimodal interventions such as surgery and/or radiotherapy treatment. The second major management dilemma of NFPMAs is the presence of hormone deficiencies which necessitates lifelong endocrinological follow-up.

Untreated non-functioning pituitary adenomas

There have been several studies looking at the behaviour of untreated non-functioning pituitary adenomas. In a systematic review of 11 studies of 865 patients, authors Fernandez-Balsells et al, found that microadenomas were unlikely to grow over time, but that the risk of growth was higher in macroadenomas (59). Macroadenomas were significantly more likely to grow over time with an incidence of 12.53 per 100 person years compared to 3.32 per 100 person years for microadenomas. Included in this systematic review are two recent European studies. In the first study, authors Karavitaki et al., reviewed 40 cases consisting of 16 microadenomas and 24 macroadenomas, reporting that only 12.5% of microadenomas grew compared to 50% of macroadenomas over a mean observation period of 41 months (60). In this study, growth was not clearly defined, but presumed to be any increase in size of the adenoma. In the group with macroadenomas that developed growth 67% of cases had a worsening or new visual field defect. The authors concluded that observation by a “watch and wait policy” alone for macroadenomas is “not a safe” option due to the high chance of visual problems. In the second study, authors Dekkers et al., included only those with macroadenomas who had either no or minor visual field defects. Adenoma growth was defined as an increase of two or more millimetres in any direction on MRI scanning. The authors also found a high prevalence of growth in 50% of cases over 118 months of observation (61). Unlike the previous study by Karavataki et al., authors Dekker et al., concluded that observation is “a safe option” in selected macroadenomas as any worsening visual field defect can be readily reversed with surgery.
In 2015, two further studies were published about the natural history of untreated non-functioning pituitary adenomas. From the United Kingdom, authors Sam et al., also took a conservative approach to the safe option of watching these adenomas despite being the first to report such high prevalence of growth in microadenomas (62). In this study, authors Sam et al., followed 19 microadenomas and 47 macroadenomas over a mean period of 4.3 years, measuring growth as any millimetre increase; they reported growth occurring in 52.6% with microadenomas and in 60% of those with macroadenomas. From Australia, authors Lenders et al, studied 27 microadenomas and 23 macroadenomas over a mean period of three years, measuring volumes with a strict definition of growth being ≥20% change (58). Increases occurred in only 7% of microadenomas but in 39% of macroadenomas. These authors found that a volumetric growth rate of ≥10mm^3/month assessed at two years for the macroadenoma group, was 90% sensitive and specific for being considered a significant growth that triggered the decision for surgical intervention (58).

The risks of complications of a conservative approach by observation of non-functioning pituitary adenomas are very low: with a new hormone deficiency occurring in 1.5 per 100 patient years (58), risk of new apoplexy being an even more rare event (58) (60) and a new onset of visual field defect can be readily reversed with surgery (60) (61) (Figure 1.9). An important management issue associated with a conservative approach for non-functioning pituitary adenomas is ensuring that patients attend for regular review as invariably some are lost to follow-up. It must be noted too that data shows that some macroadenomas can decrease in size over time, this being observed in approximately 29-34% of cases (58) (61) (62).
Figure 1.9 A, B and C. An example of a patient with a known history of a NFPM developing a sudden onset of visual disturbance due to apoplexy (63).

A: MRI coronal view depicting the adenoma contacting the under surface of the optic chiasm prior to the apoplexy occurring. B: CT scan axial view of the NFPM demonstrating haemorrhage due to apoplexy. Patchy enhancement of the pituitary mass is seen indicating haemorrhage within the adenoma. C: Visual field testing pre- and post-operatively. Pre-operatively the visual fields demonstrate absence of vision in the right eye and a dense hemianopia of the lateral visual field in the left eye, as noted by the black areas (upper panel). Post-operatively vision is restored to near normalisation, but with residual visual impairment in the right eye (lower panel).
Surgical treatment of non-functioning pituitary macroadenomas

Surgery remains the first line treatment for NFPMA. Unlike prolactinoma, medical therapy with DAs have not proven of value in the management of NFPMA.

Histologically NFPMA do express dopamine receptors and somatostatin receptors, the use of DA and somatostatin receptor ligands, respectively, have not be shown to have a significant effect on the size of NFPMA in order to be considered as first line treatment (64). Thus, surgery remains the treatment of choice and furthermore, is effective in treating adenomas causing mass effect and with a low risk of surgical complications (65). Indications for surgery for NFPMA are for symptomatic disease with mass effect, hormone deficiencies, growth during observation and pregnancy planning in pre-menopausal females (56).

Pituitary surgery for NFPMA (and other types pituitary adenomas) is usually via the transsphenoidal route, using either the microscopic technique or endoscopic technique (66). Many neurosurgical retrospective studies have been published on these two surgical techniques with both considered effective and with low risk of complications. Based on meta-analyses it is the surgeon’s expertise with either technique that is the cornerstone of importance in terms of surgical outcomes (67) (68) (69).

Even in expert neurosurgical hands, the presence of residual disease is a frequent finding post-surgery in NFPMA (Figure 1.10). Single centre series of operated cases of NFPMA throughout the world report rates of residual disease between 54-80% cases (70) (71) (72) (73) (74) (75) (76). An exceptionally low rate of residual disease of 32% was achieved by a recent Korean group using the traditional microscopic technique (77). Australian data comes from three small studies. In the first study, residual disease was found in 51% of 39 cases of NFPMA (78), in the second study in 60.5% of 38 cases (79) and in the final study in 61% of 108 cases (80). These differences in prevalence of
residual disease between centres may relate to differences in surgical expertise or in the complexity of cases of NFPMAs that present at the various centres.

The clinical problem with residual disease is that this is the largest risk factor for regrowth of NFPMAs. The rate of regrowth varies throughout the world literature. Series from Europe (70) (71) (81), Korea (77) and Israel (72) report regrowth rates between 42-59% over mean period of 4-6 years of follow-up. Notably, the lowest rates of regrowth observed over long follow-up occurred in an Irish (73), Dutch (74) and North American series (75), with regrowth of 33.5%, 14% and 28%, respectively at median times of 5.9, 5.0 and 8.4 years, respectively. A French series by authors Sotos-Ares et al., also reported low rate of regrowth of 38.3%, however this was over a short follow-up of period of mean 2.3 years (76). In an Australian series, an extremely high rate of 73% (48/66) regrew over a long median follow-up of 5.7 years (80). The reasons for such differences in reported regrowth rates are not clear, however, may relate to differences in the duration of follow-up and heterogenous definitions of regrowth. In regard to this later point, authors Ratnasingam et al., are the only group from the series presented above that have defined regrowth being a minimally detectable change of 3mm in any direction and equating to a volumetric change of 10mm³; earlier studies have not used such strict definitions (80).
Figure 1.10 A and B. A 56 year old male presented with visual disturbance and found to have a large NFPMA.

A: MRI coronal view of large NFPMA.

B: Post-operative MRI coronal view showing a small residual adenoma that has not regrown after eight years of follow-up (blue arrow).
Recurrence is defined by the return of the adenoma after complete macroscopic removal has been achieved post-operatively (Figure 1.11). The prevalence of recurrence of NFPMAs is lower than the prevalence of regrowth occurring. Series from Europe (70) (71) (81), Korea (77) and Israel (72) have reported recurrences in 6.9-20% of operated cases of NFPMAs over an average follow-up period of 4.6 years. The lowest rates of recurrence observed over long follow-up periods occurred in Irish (73), Dutch (74) and North American series (75), of 0% (0/26), 0% (0/27) and 5.2% (13/248), respectively at median times of 5.9, 5.0 and 8.4 years, respectively.

In the above series of regrowth and recurrences, most cases occur within 10 years of surgery. In a study by Reddy et al., from the United Kingdom, it was observed that 20% of recurrences occurred after 10 years follow-up, including one case at 25 years (70).

Due to this long-term concern regarding ongoing disease of NFPMAs due to regrowth and recurrence radiological follow-up is required of all cases. The timing of follow-up however remains undefined in current guidelines. Indeed, a recent systematic review and guideline was unable to conclude the optimal duration of time or frequency of follow-up for those with or without residual disease, even including those treated with radiotherapy (82). A pertinent clinical question posed by authors Reddy et al., is when to stop imaging those with operated NFPMAs who have not been treated with post-operative radiotherapy (70). These authors were unable to define a time point from their study, however, concluded that imaging should continue indefinitely and recommended that early irradiation should be considered in those with post-operative extrasellar adenomas. The French Endocrine Society’s recent consensus paper addresses the frequency of follow-up imaging, suggesting annual imaging for the first five years then every 2-3 years in a case dependent manner in those with residual disease (83). In those without residual disease, the French guidelines recommend annually imaging for the first five years, then at 7, 10, 15 years before ceasing (83).
In Chapter 3 of this thesis I will explore surgical aspects of operated cases of NFPMAs from a single Australian centre that undertakes a high volume of pituitary surgeries. This chapter will examine long-term outcomes of regrowth and recurrence and determine risk factors for ongoing disease. Despite there being much international literature on this, the literature from Australia is limited and the surgical outcomes of the local centre have not been previously examined.
Figure 1.1 A and B. A 44 year old male had a successful resection of NFPMA with no residual disease (A). He did not attend regular follow-up but represented at the age of 57 (13 years later) with symptoms related to hormone deficiencies and found to have recurrence with extensive invasion into bilateral cavernous sinuses (B). He underwent further surgery with the known plan of requiring radiotherapy given residual disease to cavernous sinus is inoperable.

A: MRI coronal view of post-operative MRI with no residual disease evident

B: MRI coronal view of extensive recurrence (blue arrow)
Management options for cases with regrowth and recurrence

Management options for cases with regrowth and recurrence of NFPMAs involve observation, repeat surgery, radiotherapy or a combination of these treatments. The complexities of these management options have been addressed in a recent American guideline specifically focused on the treatment of regrowth and recurrence of NFPMAs (84). Observation with serial MRI scanning is usually the first management approach at which time it is decided that a significant change in size of the residual or recurrent adenoma warrants additional intervention. In an Australian cohort, authors Ratnasingam et al., examined risk factors for requiring a secondary intervention. In 108 operated cases of NFPMAs with a median follow-up of 5.7 years, 22% of the cohort required a second intervention of either surgery or radiotherapy (80). In the 24 cases that required a second intervention all had residual adenoma present from the initial surgery, in comparison to those not requiring a second intervention whereby only 50% had residual disease post-operatively. The authors found that the position of the residual adenoma (in the suprasellar region) or a volumetric growth rate of >80mm³/year predicted with high sensitivity the need for secondary intervention (80).

There are no randomised studies comparing the outcomes of a second surgery to radiotherapy for NFPMAs that have regrown or recurred. The indications for each treatment modality differ and therefore decisions for optimal treatment should be individualised via multidisciplinary team consensus (83). Indications for a second surgery include adenomas causing mass effect (such as optic nerve compression), when adenoma growth is deemed to have significantly increased based on serial MRI scanning and when adenomas are deemed suitable for further resection based on location within the pituitary fossa (such that cavernous sinus adenomas are not readily accessible to surgery) (84). Repeat surgery, however, is not necessarily curative with studies reporting ongoing residual disease in 38-67% of cases undergoing a second operation due to regrowth and recurrence (85) (86) (87) (88).
Traditionally, post-operative radiotherapy was routinely administered in those with residual disease. This approach was introduced in the 1960s when it was found that most cases (75%) of NFPMAs post-surgery experienced regrowth after 10 years (89). However, over time as surgical techniques have improved, fewer cases have large residual adenomas post-surgery, thus leading to more selective cohorts receiving radiotherapy. There is no doubt that radiotherapy is highly effective at preventing further adenoma growth in most (87-100%) of cases (90). Although highly efficacious at preventing further growth radiotherapy is associated with side effects. The most well recognised adverse effect is the development of new hormone deficiencies and this has been shown to occur in at least 23% of cases at five years (90). Historically, there has also been a very small risk of radiation-induced secondary intracranial tumours (meningiomas, gliomas and sarcomas) occurring decades after radiotherapy (91). However, the risk of radiation-induced secondary malignancies appears to be minimal or negligible in more recent times where radiotherapy techniques have become precisely focused, enabling the delivery of high doses of ionizing radiation to a target whilst sparing surrounding structures of radiation exposure. For example, in a large observational study of 1837 patients over 25 years, who had been treated with intracranial single fraction radiosurgery (SRS), there were no cases of radiation-induced secondary malignancies in 11 264 patient-years observed (92). Clinical guidelines recently suggest radiotherapy can be considered in those with residual disease but with consideration of several factors including size and position of the residual adenoma, patient factors, aggressive features of the adenoma on histology and those that have demonstrated regrowth (93).

A recent study has examined the clinical outcomes of cases of NFPMAs that experienced regrowth and recurrence. With data collected from two UK centres, 237 cases with regrowth and recurrences were studied; of this cohort 94% had residual disease originally after their first operation and the remaining 6% had no residual disease (94). Management of the first episode of regrowth and recurrence in the 237 cases included observation in 40.3% and an intervention in the remaining 59.7% (further surgery in 14%, radiotherapy in 24.6%, or both modalities in 21.2%). During a
median follow-up of 5.9 years, 90 cases had a “second” regrowth; this predominately occurred in those who were being observed (63.4% of cases observed) rather than those cases that had an intervention (37.6% of cases with an intervention). Of the cases managed by observation alone, a third were eventually offered an intervention (surgery or radiotherapy or a combination) because of further enlargement. Lastly, there were even a small number of cases that developed a “third” regrowth, with one case that transformed into a pituitary carcinoma. Radiotherapy was most effective at preventing repeated episodes of regrowth. These data highlight the long-term threat of persistent disease state in those with NFPMAs.

There has been increased interest in the use of medical therapy of the DA cabergoline in preventing regrowth of those with residual disease. D2 receptors are variably expressed in non-functioning adenomas (95) (96). Six observational studies (totalling 109 patients) have demonstrated that giving cabergoline in variable doses between 1.0- 3.0mg weekly to those with residual disease, that only 14% of cases demonstrated regrowth and 42% of cases demonstrated volumetric shrinkage of 10-25%. This is in contrast to the rates of regrowth in approximately 40-50% of cases not treated with cabergoline. In four of these studies the observation period of follow-up was relatively short between 6-12 months (96) (97) (98) (99), but in the remaining two studies the observation periods were longer at 5 and 6.5 years (100) (101). Although only a limited number of patients have been studied so far, the results regarding the potential of cabergoline to attenuate adenoma regrowth looks promising and warrants further testing in a randomised controlled trial.

**Hormone deficiencies**

NFPMAs are associated with hormone deficiencies pre- and post-operatively, occurring with greater frequency compared to secretory adenomas and microadenoma (102), (103) (104) (105) (106) and when NFPMAs are resected trans-cranially compared to the
trans-sphenoidal route, likely due to trans-cranially operated cases having much larger adenomas (66) (107).

There is a hierarchy of hormone deficiencies which occur in NFPMAs with growth hormone and gonadotropin deficiencies occurring in 85% and 75% of cases respectively, and with fewer deficiencies in the ACTH-cortisol axis and TSH axis, with 38% and 32% of cases, respectively (74) (107) (108) (109) (110) (111). The mechanisms of hormone deficiencies relate to a complex interplay between adenoma size, compression of the pituitary stalk and impaired signalling from the hypothalamus and pituitary gland, and compression and destruction of normal pituitary tissue by the adenoma. Prolactin, however, is the one hormone that can be elevated in NFPMAs due to the “stalk effect”; although it has been shown that the maximum limits of prolactin in cases with NFPMAs rarely exceed 2000mIU/L (29). The “stalk effect” is commonly seen in NFPMAs occurring in 38.5-44.8% of cases (20) (112). Apoplexy is associated with a high prevalence of hormone deficiencies, including prolactin deficiency which seldom occurs in non-apoplectic NFPMAs.

Post-operatively, pre-existing hormone deficiencies can persist, improve or new hormonal axis deficiencies can develop. Historically, author Baha Arafah and colleagues have written extensively on hormonal outcomes in cases of NFPMAs. In one study, of 26 cases of large NFPMAs (of mean maximum diameter of 31mm), hormone deficiencies at presentation occurred with the following frequencies: 100% with growth hormone deficiency, 96% (25 cases) with gonadotropin deficiency, 81% (21 cases) TSH with deficiency and 62% (16 cases) with ACTH-cortisol deficiency (106). Post-surgery, only 15% of cases recovered from growth hormone deficiency, but recovery in the other axes were seen in 32-57%. The authors observed that hormonal recovery was more likely in those with higher prolactin levels at baseline and those with smaller adenomas (<25mm in maximum diameter), proposing that these features were markers of preserved and viable pituitary tissue (106).
In a further work, Arafah et al., examined 35 cases with ACTH-cortisol deficiency prior to surgery, again finding that those with an elevated prolactin were more likely to recover the ACTH-cortisol axis (110). This supported their hypothesis that hypopituitarism due to compromise of the pituitary venous delivery system by the adenoma (signalled by raised prolactin level) was a reversible process, so long as there was viable (non-necrosed) pituitary tissue post-operatively.

Several years later, Arafah et al., continued to explore hypopituitarism and relationship to prolactin levels with another variable of pressure within the adenoma called intrasellar pressure (ISP) (113). The authors found that higher ISP positively correlated with both hypopituitarism and those with higher prolactin levels at presentation. They concluded that ISP has a crucial role in the mechanism of development of hypopituitarism and hyperprolactinaemia. On page 80 of this literature review I expand on the topic of ISP and its relationship with pituitary adenoma size and hormone function.

In Chapters 4 and 5 of this thesis I will explore hormonal aspects of NFPMA. In Chapter 4, I describe a large Australian cohort of operated cases of NFPMA examining gender differences and long-term hormonal outcomes. In Chapter 5, I will examine the role of ISP and its relationship to hormonal outcomes and adenoma size in operated cases of NFPMA.
1.5.3 Somatotropinomas

Somatotropinomas secrete excessive growth hormone (GH). This leads to the clinical syndrome known as acromegaly in adults and gigantism in children if it occurs prior to the fusion of the bone epiphyses. Somatotropinomas can also cause problems due to mass effect when adenomas are large. The hypersecretion of GH signals a rise in insulin-like growth factor 1 (IGF-1) from the liver, which has the peripheral effects of altered metabolism and increased growth. Raised GH and IGF-1 leads to the multisystem diseases of hypertension, diabetes, sleep apnoea, arthritis and changes to the physical appearance (Figure 1.12). Complications of untreated acromegaly are the increased mortality due to cardiovascular and respiratory causes and morbidity of arthropathy. In the last decade, there is evidence that mortality due to cardiovascular and respiratory appears to be decreasing, but superseded by deaths due to malignancies, similar to the background population (114). This decrease in mortality is likely due to the widespread use of adjuvant medical therapy (somatostatin receptor ligands) in lowering GH and IGF-1 levels. Lowering the GH and IGF-1 levels to within the normal reference range has been shown to reduce mortality to that of the background population (114) (115).

The physical changes that occur in acromegaly are insidious in onset and can take approximately five years to diagnose. One study has examined the signs and symptoms of acromegaly over the period of 1980-1994 compared to 1995-2006 finding that the diagnosis of acromegaly still took a mean of 5.9 years from the first onset of patient symptoms despite the passage of time (116). In the 324 cases described the commonest symptoms that led to the diagnosis of acromegaly were changes in physical appearance (facial and/or enlargement of hands and feet) in 22% of cases; these changes in appearance were more likely to be noted by others rather than the individual patient, thus highlighting the insidious nature of acromegaly. Changes in vision was the second commonest symptom to prompt diagnosis which occurred in 18% of cases (116).
Figure 1.12 A, B, C and D. Typical features of acromegaly in a 55 year old female. A: enlarged hand (left) compared to an adult female (right), B: markedly enlarged feet, C: enlarged facial features, thickening of skin and macroglossia, D: multiple skin tags around neck (photos obtained with patient permission).
Although the time to diagnosis is still relatively long, the situation has improved since historical descriptions of acromegaly in 1890s, whereby diagnosis could take up to 10-20 years (117). The signs and symptoms described in the early cases were of profound changes that included bone deformation, blindness, deafness, severe rheumatism, muscle wasting, polyuria (likely due to diabetes mellitus) and fractures of the thoracic spine causing significant deformity (117). In fact, fractures of the thoracic spine have been recently redescribed with a meta-analysis reporting a median prevalence of 40% of acromegalic patients with radiological vertebral fractures, which is three to eight fold greater than control subjects (118). The mechanisms of this increased prevalence of fractures are still being elucidated but can be partly explained by the findings of elevated bone turn over makers and bone formation markers (but to a lesser degree) in acromegalic patients resulting in an increase in bone fragility (118).

The aims of treatment of somatotropinomas are to normalise both GH and IGF-1 levels and to reduce mass effect of the adenoma. There are several treatment options possible to achieve these goals including surgery, medical therapy and radiotherapy. Surgery is considered and recommended as first line therapy and is the only treatment that can achieve immediate cure (119) (120). With somatotropinomas, the likelihood of biochemical remission post-operatively is related to adenoma size, with remission being more likely in those with smaller adenomas (>70%) compared to those with macroadenomas (50%) (121) (122). Remission is also dependent on surgical expertise (78) (121) (123) (124) (125) (126) and has improved in recent times due to better surgical techniques, additionally as adenomas appear to be diagnosed sooner in the disease process with smaller adenomas (127). Approximately 40% of patients with acromegaly do not achieve biochemical remission post-operatively, necessitating additional treatment options such as medical therapies, combination of medical therapies and radiotherapy (128). In those who have achieved biochemical remission post-operatively the risk of recurrent disease is low with one large series reporting a long-term follow-up of 15 years with only four recurrences in 61 patients (6.5%) occurring at 4, 7, 8 and 12 years respectively for each case, after surgery (127).
There are several medical therapies in the armamentarium for treating acromegaly. The most established of these medical therapies are the first generation somatostatin receptor ligands (SRLs); these agents are agonists at the somatostatin receptors (particularly receptor subtypes 2 and 5) leading to inhibition of GH secretion from the pituitary gland. First generation SRLs are effective at reducing both GH and IGF-1 levels in 17-70% of cases in clinical studies (129). As there is such a variable response to first generation SRLs a second generation SRL was developed with high binding affinity to somatostatin receptor subtype 5: it can normalise GH and IGF-1 levels in an extra 20% of cases resistant to first generation SRLs (129). Another of the medical therapies is GH antagonist pegvisomant. It is a highly selective GH antagonist at the level of the liver and can reduce IGF-1 levels in 97% of cases in clinical trials but in approximately 60% in the real world experience (129). Pegvisomant is often used in combination with SRLs in other parts of the world (130), however prescribing criteria outlined by the Pharmaceutical Benefits Scheme (PBS) in Australia, only permits the use of pegvisomant as monotherapy. Lastly, the DA cabergoline (also used for prolactinomas) has shown some limited benefit as single agent or combination with the other medical therapies for acromegaly with better responses to treatment in those patients with near normal IGF-1 levels (131). A meta-analysis on the use of cabergoline in acromegalic patients showed that one third of cases achieved biochemical control using cabergoline as a single agent and an additional 50% achieved biochemical control when cabergoline was combined with SRLs in those not controlled with SRLs alone (131).

There are many options of medical therapies for treating acromegaly and traditionally the use of these medications have been via a step by step approach particularly in Australia due to prescribing limitations, as mentioned above. However, it has been suggested by one group of authors that the use of these medical therapies in acromegaly should be “personalised” based on biomarkers such as histological staining pattern of somatostatin receptor subtypes and dopamine receptors (132).
Given such complexities in the management of acromegaly, it remains that a multidisciplinary team approach is recommended for optimal patient care (120).
1.5.4 Corticotropinomas (Cushing’s Disease)

Corticotropinomas are pituitary adenomas that secrete excessive adrenocorticotrophic hormone (ACTH). The abnormal production of ACTH results in excessive cortisol production from the adrenal glands which in turn leads to the clinical condition known as Cushing’s disease (CD). Excessive cortisol production causes hypertension, metabolic diseases of diabetes and hyperlipidaemia, myopathy, cardiac failure, osteoporosis and fractures and an increased susceptibility to infections. The physical manifestations of CD include rounded facies, central adiposity, thin skin and large striae (Figure 1.13). Left untreated the effects of hypercortisolaemia leads to significant co-morbidities and reduced life expectancy (133). It has been appreciated more recently that hypercortisolaemia can cause negative effects on brain function due to the presence of glucocorticoid receptors. These effects result in cognitive issues such as depression, anxiety, reduced concentration and sleep disturbance; these effects may not be fully reversed in those achieving disease remission and can be a major cause of morbidity (133).

Primary treatment for corticotropinomas is by pituitary surgery. Most of these adenomas are microadenomas (Figure 1.14). Similar to somatotropinomas, corticotropinomas are problematic as remission is less likely in those with macroadenomas compared to microadenomas. In a meta-analysis of surgical outcomes in those with CD 83% of those with microadenomas and only 63% with macroadenomas were in remission: an overall remission rate of 76% (134). For those patients who have been in biochemical remission from their first surgery for corticotropinomas there is a well-known risk of disease recurrence as high as 15–66% in various long-term studies (135); this high risk of recurrence of CD necessitates lifelong follow-up of all cases. In those with persistent or recurrent disease there are several treatment options that can be considered by the multidisciplinary team including repeat pituitary surgery, medical therapies, pituitary radiotherapy and bilateral adrenalectomy (135). There are numerous medical therapies than can be used to treat ongoing hypercortisolaemia which either target the pituitary directly to lower
ACTH levels (such as SRLs) or lower cortisol production at the level of the adrenal glands. Many of these medical agents are difficult to administer due restrictions on availability and limitations due to numerous side effects. Recently, the DA cabergoline has also been studied as a pituitary directed treatment showing some limited benefit when used as either single treatment or in combination with other medical therapies (136).

Corticotropinomas are the most complex of pituitary adenoma types. From diagnosis, to the various treatment options and the long-term management of its complications, hence the need for a multidisciplinary team approach to treatment decision making.
Figure 1.13 A and B. Typical features of hypercortisolaemia in a 23 year old female with a confirmed corticotropinoma. She presented with headaches and long-standing history of weight gain. Her features are in fact subtle in appearance.

A: Patient has a rounded facies and suprACLavicular fat pads, B: central adiposity with abdominal striae (photos obtained with patient permission).

Figure 1.14 A and B. Post-contrast MRI shows left sided microadenoma in the above patient (blue arrow). She underwent pituitary surgery with confirmed an ACTH staining adenoma on immunohistochemistry. The patient continues to be in biochemical remission of CD.

A: MRI coronal view  B: MRI sagittal view
1.5.5 Thyrotropinomas (TSHomas)

Thyrotropinomas (TSHomas) secrete excessive thyroid stimulating hormone (TSH) and α-glycoprotein subunit. Over 450 cases have been described in the literature with the first cases being reported in 1960 having hyperthyroidism and an enlarged sella on skull x-ray with remission achieved by pituitary radiotherapy (137) (138). In the presence of a TSHoma, TSH levels can be elevated or inappropriately normal and lead to various degrees of hyperthyroidism; some cases are clinically silent. Historically, cases with hyperthyroidism due to a TSHoma have been mistaken for Graves’ disease. TSHoma can also co-secrete growth hormone and prolactin with this can occurring in 16% and 10-12% of cases respectively (139) (140).

TSHomas in most series are more likely to present as macroadenomas compared to microadenomas (Figure 1.15). Presentation can be incidental, due to mass effects or due to hypersecretion of TSH or co-secreted hormones (140) (141) (142).

TSHomas have been shown to respond to medical therapy with SRLs which can lead to hormonal control and adenoma shrinkage. In one large series of 90 TSHomas in a single centre, euthyroidism was achieved in 40/48 (83%) of patients and adenoma shrinkage was found in 24/44 (55%) of patients following pre-operative SRL treatment (140).

Surgery is the best definitive management of TSHomas. In this series above, surgical treatment remission was achieved overall in 76 (84%) of 90 patients, 100% cases with microadenomas and 81% of cases with macroadenomas. There were no cases of adenoma recurrence during a median follow-up period of 2.8 years (140). In another smaller study recurrent TSHomas, after complete resection, was also rare with no recurrences in 14 cases after a mean of 7.5 years of follow-up (143). Cases not
achieving biochemical normalisation or adenoma mass control have been treated with a combination of SRLs and radiotherapy (140).
Figure 1.1 A and B. A 32 year old male presented with lassitude and gynecomastia. Endocrine investigation revealed low cortisol and testosterone levels explaining his symptomatology. The TSH level was mildly elevated with normal thyroid hormone levels and raised alpha subunit. MRI scanning demonstrated a large macroadenoma elevating the optic nerve superiorly yet surprisingly he did not have any visual disturbance (A). The working diagnosis was that the patient has a TSHoma and this was confirmed post-operatively on immunohistochemical staining of the adenoma. Post-operatively, the patient has a small residual adenoma and has cortisol and growth hormone deficiencies, but he has normal gonadal function.

A: MRI coronal view of a 22 x21x 30mm adenoma (blue arrow)

B: MRI sagittal view of the adenoma (blue arrow)
1.5.6 Gonadotropinomas

Gonadotropinomas are adenomas that have traditionally been considered a subtype of non-functioning adenomas. They are characterised histologically by immunohistochemical staining for β-FSH, β-LH or α-subunit and more recently by staining for steroidogenic factor-1 (SF-1). In one surgical series of operated cases of NFPMA, hormonally silent gonadotropinomas accounted for 64% of cases (144). A small subset of gonadotropinomas are considered functional as the secretion of biologically active hormone (usually FSH) results in the clinical syndrome of ovarian hyperstimulation in pre-menopausal females due to raised estrogen levels or testicular enlargement and raised testosterone in males (145) (146). These adenomas are generally large and require surgical removal. Given the rarity of these adenomas there are no large case series describing the long-term recurrence rates (147).
1.5.7 Updated classification of pituitary adenomas

In 2004, the WHO classification of pituitary adenomas categorised these as either typical, atypical and pituitary carcinomas. Most adenomas are considered typical based on their indolent clinical behaviour and having evidence of low proliferative indices on histological analysis (markers such as Ki-67 and p53). Atypical adenomas tend to behave more aggressively, are invasive, more treatment resistant, associated with higher proliferative indices on histology and likely to recur. Atypical adenomas are found in 2.7-15% of cases of pituitary adenomas (148) (149). Pituitary carcinomas are those that disseminate in the cranio-spinal space or demonstrate systemic spread and occur in 0.2% of cases (150).

Do to advances in the understanding of the pathophysiology of pituitary adenomas since 2004, pituitary adenomas have been further classified based on a combination of traditional histological hormone staining with an additional description of morphological changes (such as patterns of granulation-sparse and dense in somatotropinomas) and by the presence of transcription factors and co-markers. These changes have been incorporated in the 2017 4th edition WHO classification of pituitary tumours (13) (Table 1.2).

From a clinical perspective, the new classification is of some clinical utility. For example, categorising somatotropinomas into densely and sparsely granulated is of clinical utility in predicting response to medical treatment with first- and second-generation SRLs (151). This new classification has further categorised non-functioning adenomas into gonadotrophic, plurihormonal and pure null cell adenomas based on differences in immunohistochemical staining and by presence of certain transcription factors, although at this stage the clinical significance of this subdivision of non-functioning pituitary adenomas is not entirely clear.
Table 1.2 2017 WHO classification of pituitary adenomas (modified from WHO classification of tumours of the pituitary gland) (13).

<table>
<thead>
<tr>
<th>Adenoma type</th>
<th>Morphological variants</th>
<th>Pituitary hormones and other immunomarkers</th>
<th>Transcription factors and other co-factors</th>
</tr>
</thead>
<tbody>
<tr>
<td>Somatotroph adenomas</td>
<td>Densely granulated adenoma</td>
<td>GH ± PRL ± α-subunit</td>
<td>PIT-1</td>
</tr>
<tr>
<td></td>
<td>Sparsely granulated adenoma</td>
<td>GH ± PRL, [CK]</td>
<td>PIT-1</td>
</tr>
<tr>
<td></td>
<td>Mammosomatotroph adenoma</td>
<td>GH + PRL (in same cells) ± α-subunit</td>
<td>PIT-1, ERα</td>
</tr>
<tr>
<td></td>
<td>Mixed somatotroph–lactotroph adenoma</td>
<td>GH + PRL (in different cells) ± α-subunit</td>
<td>PIT-1, ERα</td>
</tr>
<tr>
<td>Lactotroph adenomas</td>
<td>Sparsely granulated adenoma</td>
<td>PRL</td>
<td>PIT-1, ERα</td>
</tr>
<tr>
<td></td>
<td>Densely granulated adenoma</td>
<td>PRL</td>
<td>PIT-1, ERα</td>
</tr>
<tr>
<td></td>
<td>Acidophilic stem cell adenoma</td>
<td>PRL, GH (focal and variable)</td>
<td>PIT-1, ERα</td>
</tr>
<tr>
<td>Thyrotrhop adenoma</td>
<td></td>
<td>β-TSH, α-subunit</td>
<td>PIT-1</td>
</tr>
<tr>
<td>Corticotroph adenoma</td>
<td>Densely granulated adenoma</td>
<td>ACTH, [CK]</td>
<td>T-PITb</td>
</tr>
<tr>
<td></td>
<td>Sparsely granulated adenoma</td>
<td>ACTH, [CK]</td>
<td>T-PITb</td>
</tr>
<tr>
<td></td>
<td>Crooke’s cell adenoma</td>
<td>ACTH, [CK]</td>
<td>T-PITb</td>
</tr>
<tr>
<td>Gonadotroph adenoma</td>
<td></td>
<td>β-FSH, β-LH, α-subunit (various combinations)</td>
<td>SF-1, GATA2, ERα</td>
</tr>
<tr>
<td>Null cell adenoma</td>
<td>None</td>
<td></td>
<td>none</td>
</tr>
<tr>
<td>Pluri-hormonal adenoma</td>
<td>Pluri-hormonal PIT-1 positive adenoma</td>
<td>GH, PRL, β-TSH ± α-subunit</td>
<td>PIT-1</td>
</tr>
<tr>
<td></td>
<td>(previously called silent subtype 3 adenoma)</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Adenomas with unusual immunohistochemical combinations</td>
<td>Various combinations: ACTH/GH/ACTH/PRL</td>
<td>N/A</td>
</tr>
</tbody>
</table>
1.6 Frequency of pituitary adenomas

The commonest types of pituitary adenomas in the community are prolactinomas and non-functioning adenomas, collectively accounting for approximately 80% of adenomas (Table 1.3). This is followed by somatotropinomas and corticotropinomas accounting for approximately 15%; TSHomas are rare and usually account for <1% of adenomas (152). Epidemiological data for functioning gonadotropinomas are scarce, with these adenomas usually being described in case reports.

Table 1.3 Types of pituitary adenomas in community-based studies

<table>
<thead>
<tr>
<th>Location</th>
<th>Prolactinoma</th>
<th>Non-functioning adenoma</th>
<th>Somatotropinoma</th>
<th>Corticotropinoma</th>
<th>Thyrotropinoma</th>
<th>Unspecified</th>
</tr>
</thead>
<tbody>
<tr>
<td>Leige Belgium (18)</td>
<td>66%</td>
<td>14.7%</td>
<td>13.2%</td>
<td>5.9%</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Oxford UK (20)</td>
<td>57%</td>
<td>28%</td>
<td>11%</td>
<td>2%</td>
<td>-</td>
<td>2%</td>
</tr>
<tr>
<td>Fribourg Switzerland (153)</td>
<td>57%</td>
<td>29.5%</td>
<td>9%</td>
<td>4.5%</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Malta (154)</td>
<td>46.2%</td>
<td>34.2%</td>
<td>16.5%</td>
<td>2.2%</td>
<td>0.94%</td>
<td>-</td>
</tr>
<tr>
<td>Western Sweden (155)</td>
<td>32%</td>
<td>54%</td>
<td>9%</td>
<td>4%</td>
<td>0.7%</td>
<td>-</td>
</tr>
<tr>
<td>Iceland (31)</td>
<td>39.9%</td>
<td>43%</td>
<td>11.3%</td>
<td>5.7%</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td>Total range</td>
<td>32-66%</td>
<td>14.7-54%</td>
<td>9-16.5%</td>
<td>2-5.9%</td>
<td>&lt;1%</td>
<td>-</td>
</tr>
</tbody>
</table>
1.7 Prevalence of pituitary adenomas

Pituitary adenomas have traditionally been considered uncommon, however more recent community-based population studies show that clinically relevant adenomas occur in approximately 1 per 865-1470 (Table 1.4). In Australia, with a population 24 million, this would equate to approximately 24,000 cases having a clinically relevant pituitary adenoma. Prolactinomas and non-functioning adenomas are the most prevalent pituitary adenoma type to occur.

As shown in Table 1.4 below, there is evidence that the prevalence of pituitary adenomas appears to be increasing over time, with most notable increases in prevalence of prolactinomas and non-functioning adenomas. The most recent of these studies, originating from Iceland with a homogenous population and a single health care system, the prevalence of pituitary adenomas was 1 per 865 population. These authors compared pituitary adenoma prevalence over time with an impressive increase from 6.63/100 000 in 1972 to 115.57/100 000 in 2012 (31). The authors also observed that the increase in prevalence over time occurred due to an increase in cases of prolactinomas and non-functioning adenomas, rather than in cases of somatotropinomas and corticotropinomas (31).
<table>
<thead>
<tr>
<th>Location</th>
<th>cases/100,000 population</th>
<th>1 Case per population</th>
<th>Adenoma type/ 100,000</th>
</tr>
</thead>
<tbody>
<tr>
<td>Stoke on Trent, UK 1988-1998 (156)</td>
<td>19-28</td>
<td>1 per 3571-5263</td>
<td>PRL 6-10 NFA 7-9 GH 4-6 ACTH 2-3</td>
</tr>
<tr>
<td>Leige, Belgium (18)</td>
<td>94</td>
<td>1 per 1064</td>
<td></td>
</tr>
<tr>
<td>Fribourg, Switzerland (153)</td>
<td>80.5</td>
<td>1 per 1241</td>
<td>PRL 45.8 NFA 23.8 GH 7.3 ACTH 3.7</td>
</tr>
<tr>
<td>Oxford, Uk (20)</td>
<td>77.6</td>
<td>1 per 1288</td>
<td>PRL 44.4 NFA 22.2 GH 8.6 ACTH 1.2 Unspecified 1.2</td>
</tr>
<tr>
<td>Northern Finland (157)</td>
<td>68</td>
<td>1 per 1470</td>
<td></td>
</tr>
<tr>
<td>Malta (154)</td>
<td>75.7</td>
<td>1 per 1321</td>
<td>PRL 35 NFA 36 GH 12.5 ACTH 1.7 TSH 0.9</td>
</tr>
<tr>
<td>Iceland (31)</td>
<td>115.57</td>
<td>1 per 865</td>
<td>PRL 54.37 NFA 42.32 GH 13.67 ACTH 6.21</td>
</tr>
<tr>
<td>Overall Range from 2006-2015</td>
<td></td>
<td>1 per 865-1470</td>
<td></td>
</tr>
</tbody>
</table>

PRL: prolactinomas, NFA: non-functioning adenomas, GH: somatotropinomas, ACTH: corticotropinomas, TSH: TSHomas
1.8 Incidence of pituitary adenomas

Data from community-based studies also appears to show an increasing incidence of pituitary adenomas over time, with an overall incidence 3.98-4.8/100,000/year (Table 1.5). The most recent study by authors Agusston et al., shows the highest standardised incidence rate of 5.8/100,000/year (31). In this study, the incidence of prolactinoma and non-functioning adenomas had increased over the time periods studied; this study also showed a major increase in prevalence of prolactinoma and non-functioning adenomas, as described in the previous section of this literature review. The authors of this study have hypothesised that this increase in both incidence and prevalence is not a random observation, but partly explained by the introduction of MRI scans in the 1990’s and also due to increased awareness of symptoms with the appropriate use of diagnostic tests such as MRI scans given the majority of cases presented with symptoms related to their pituitary adenomas (and without an increased frequency of incidentally discovered adenomas) (31).

In Australia, population 24 million, the expected incidence of pituitary adenomas may be 1329 new cases per year, with approximately 1000 new cases of prolactinomas and non-functioning occurring adenomas annually.

These data, demonstrating the increasing prevalence and incidence rates of pituitary adenomas (and specifically prolactinomas and non-functioning adenomas), highlights the importance of addressing clinical aspects of these conditions that are relevant to these patient groups; hence the purpose of my thesis.
### Table 1.5. Incidence of pituitary adenomas in community-based studies

<table>
<thead>
<tr>
<th>Location</th>
<th>Year</th>
<th>Overall SIR/ 100,000/ yr</th>
<th>Standardised incidence rate by adenoma type/ 100,000/ yr</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sweden (158)</td>
<td>1958-1991</td>
<td>0.6 1.1</td>
<td>-</td>
</tr>
<tr>
<td>Stoke on Trent, UK (156)</td>
<td>1988-1998</td>
<td>-</td>
<td>PRL 0.6-1.0  NFA 0.7-0.9  GH 0.4-0.6  ACTH 0.2-0.3</td>
</tr>
<tr>
<td>Northern Finland (157)</td>
<td>1992-2007</td>
<td>3.98</td>
<td>PRL 2.16  NFA 1.02  GH 0.34  ACTH 0.17</td>
</tr>
<tr>
<td>Malta (154)</td>
<td>2000-2011</td>
<td>4.27</td>
<td>PRL 2.05  NFA 1.79  GH 0.31  ACTH 0.07  TSH 0.04</td>
</tr>
<tr>
<td>Sweden (159)</td>
<td>1987-2011</td>
<td>5.8</td>
<td>PRL Males 1.4  Females 3.6  NFA Males 2.6  Females 2.2  GH Males 0.8  Females 0.4  ACTH Males 0.3  Females 0.5</td>
</tr>
<tr>
<td>Iceland (31)</td>
<td>2003-2012</td>
<td>5.8</td>
<td>PRL Males 1.4  Females 3.6  NFA Males 2.6  Females 2.2  GH Males 0.8  Females 0.4  ACTH Males 0.3  Females 0.5</td>
</tr>
</tbody>
</table>

PRL: prolactinomas, NFA: non-functioning adenomas, GH: somatotropinomas, ACTH: corticotropinomas, TSH: TSHomas
1.9 Gender and age distribution of pituitary adenomas

There are differences in gender and age distribution of pituitary adenomas.

Prolactinomas have a peak incidence between the ages of 20-40 years, with a female preponderance (160). This gender difference is probably related to the earlier presentation of pre-menopausal females presenting sooner in the natural history of the disease due to menstrual irregularities caused by hyperprolactinaemia (27) (31). After the age of 50 years, prolactinomas occur equally in males and females (27).

Non-functioning adenomas have a peak incidence from the 4-8th decade and occur in both females and males similarly (55).

In acromegaly the median age of diagnosis occurs in the 5th decade with most studies showing equal sex distribution. Cases of acromegaly can occur in the very young and in one study 22% of cases occurred in those 0-19 years (18). Historically, in an early description of acromegaly over a century ago in 1891, the age distribution of acromegaly was also noted with cases occurring either in adolescence or after the age of 30 years (117).

Corticotropinomas predominately occurs in females with peak incidence in those 35–44 years of age. In males, highest incidence occurs in those 55–64 years of age (161).

TSHomas and gonadotropinomas are the least common adenoma types and epidemiological data is scarce. In the largest population-based study of TSHomas it was found that there was a female preponderance and with a peak incidence occurring between the ages of 55-69 years in Sweden (152).
1.10 Australian admissions data

Australia has a medical system provided by a public and private sector. This can be a major barrier for collecting national pituitary data. One recent group have published the first Australian data for admissions and pituitary surgeries throughout the country, derived from a national registry for all hospital admissions, of both the public and the private sector (162). Examining data from 2000-2015, the average number of pituitary surgeries for pituitary adenoma was 31.6/ million/ year and they demonstrated a 35.6% increase over this time period. Surgeries were predominately for non-functioning pituitary adenomas (76.3%), acromegaly (21.3%), Cushing’s disease (1.9%) and other types of adenomas (0.5%). Over the time period examined, there was significant increase in admissions for non-functioning adenomas from 29.8/ million/year to 40.6/ million/ year, however these increases were not reflected in other types of adenomas. Most pituitary surgeries (70%) occurred in those aged over 45 years with highest rates occurring in those aged 55-64 years (162).
1.11 Quality of life in patients with pituitary adenomas

Quality of life (QOL) in medicine can be defined as “the functional effect of an illness and its consequent therapy upon a patient, as perceived by the patient” (163). It is measured objectively by questionnaires, which can be either general or disease-specific and cover aspects of social, psychological and physical functioning. Of recent times interest in the quality of life of those with pituitary adenomas has increased. In a recent review, based on 102 studies up until January 2014, found that each year over the last 3 decades has seen a steady rise in published studies of QOL in pituitary diseases (164). Between 2001-2003, there were 1-3 studies published in each year, compared to 2011-13 where there were 11-14 articles published. In this review of 102 studies, 49 different questionnaires were used, making it difficult to compare study groups. The commonly used questionnaires for pituitary adenomas were disease-specific questionnaires combined with a general or domain-specific questionnaire. Disease-specific questionnaires are available for acromegaly, Cushing’s disease, hypopituitarism and growth hormone deficiency. The Short Form-36 (SF-36) questionnaire is one of the commonest of the generic QOL questionnaires used in numerous studies and medical conditions, especially in publications regarding pituitary diseases (164). Being a generic questionnaire, it captures the burden of the primary condition and that of comorbid conditions, which makes it a popular tool.

Meta-analyses examining QOL in those with pituitary adenomas consistently show that QOL is affected in all types of pituitary adenoma (non-functioning pituitary adenoma, prolactinoma, acromegaly and Cushing’s disease (CD)) with significantly worst QOL reported in those with CD and acromegaly (164) (165). In those with active CD and acromegaly QOL is particularly affected as there is a high prevalence of psychiatric disorders with depression, psychosis and anxiety reaching 77% in those with CD and 63% in those with acromegaly (165). The influence of specific treatments of pituitary adenomas on QOL, such as hormonal replacement and radiotherapy, is not particularly clear as there is much heterogeneity in the literature in regards to these factors: some
studies report that hormone deficiency and radiotherapy can reduce QOL, yet not all studies have found this (166) (167) (168).

A newly developed pituitary-specific questionnaire has been developed, the Leiden Bother and Needs Questionnaire (LBNQ-Pituitary) (169) (170). Created by analysing the responses of a focus group of patients with different types of pituitary disorders discussing issues affecting them, it was developed to be academically useful, but also clinically useful as it addresses mood problems, negative illness perceptions, issues in sexual functioning, physical and cognitive complaints, and issues in social functioning. The authors have compared this questionnaire to six other validated questionnaires (EuroQol-5D, SF-36, MFI-20, HADS, AcroQol, Cush-ingQoL) and found that there was very good reliability. Of interest, their data showed certain issues related to the specific type of pituitary adenoma. Those with non-functioning adenomas were more likely to experience problems related to impaired vision, those with prolactinoma had more negative thought perceptions of medications, and those with Cushing’s disease were more likely to report cognitive issues such as sleeping problems, difficulty with letting go of thoughts and feelings of sadness, shame and jealousy. Further data demonstrated that pituitary patients were bothered by fatigue (17%), had difficulty in performing work (12%), had problems concentrating (11%) and were less interested in sex (10%) (170).

The burden of disease experienced by pituitary patients is significant and reflective in the day to day clinical management of these cases. Understanding this psychological burden is an important part of management and considered part of patient education and support in the multidisciplinary care of cases with pituitary adenomas (165) (171). Recently this has even been addressed in guidelines for the management of Cushing’s disease, to consider psychiatric and psychological needs of these cases (135).
1.12 The need for multidisciplinary care of patients with pituitary adenomas

From the information presented thus far in this literature review, it should be evident that the management of patients with pituitary disease is complex. Appreciating that the incidence and prevalence rates of pituitary adenomas appear to be increasing (as demonstrated in Tables 1.4 and 1.5) and that the study of pituitary adenomas is expanding rapidly (ie. in discovering new genetic abnormalities, changes in classification of pituitary adenomas and navigating complex therapeutic medicines and combination treatments), is therefore understandable that there is an urgent call for pituitary patients to be optimally managed in centres that can offer multidisciplinary care. Authors have called for the involvement of a “multidisciplinary team” with particular focus on the endocrinologist for managing and co-ordinating these issues (172) (173) and furthermore, that endocrinologists have a duty of care to refer to the “best” surgeons for optimal patient outcomes (174). In terms of surgical outcomes (in those requiring operative management), there is increasing evidence that firstly, surgery should be undertaken by specialist neurosurgeons who regularly undertake pituitary surgery as there are fewer complications due to the higher volume of expertise (175). Secondly, that the remission rate of functioning adenomas (especially acromegaly) are higher in centres that have specialist pituitary surgeons (121) (123) (124), and lower remission rates where multiple neurosurgeons operate (176) (177) (178). Thirdly, surgical expertise increases over time by performing the same procedure (78) (125) (126).

A group of American endocrinologists and neurosurgeons have even proposed that the practice of pituitary diseases head towards the development of “pituitary centres of excellence”, whereby the major aims of the centre are to provide comprehensive care and support to patients with pituitary disorders, provide doctor training, continuing medical education in the management of pituitary and neuroendocrine disease and contribution to research in pituitary disorders (171). This has now been endorsed by
The Pituitary Society (an international organisation focused on the study of the pituitary gland) in their 2017 position statement (179).
1.13 Intrasellar pressure

The potential interaction between intrasellar pressure (ISP) and pituitary hormone deficiencies has already been introduced earlier in this review on page 55. ISP is the pressure within the pituitary gland as it sits in the sellar turcica. In order to understand ISP an understanding of intracranial pressure (ICP) is required. ICP refers to the pressure within the brain parenchyma and cerebrospinal fluid as it sits within the calvarium. ICP has a relationship to perfusion pressure of the brain, in that arterial perfusion needs to exceed that of ICP so that the brain receives adequate blood supply for function and survival; such that cerebral perfusion pressure (CCP) = mean arterial pressure – ICP (180). A rise in ICP is pathological as it impairs cerebral perfusion and consequently impairs brain function. This is usually seen particularly in the setting of severe brain trauma and subarachnoid haemorrhage.

The measurement of ICP was first described in 1951 where measurements were taken to measure intraventricular pressure in several pathologies using an electronic transducer (181). Since then, the procedure has been refined using fibreoptic catheters. ICP is measured by the placement of a very small pressure probe traversing the calvarium into the brain parenchyma or intraventricular space and used for management decision and prognostication of acute brain injury. In acute brain injury, a raised ICP leads to compromised cerebral blood flow, and therefore causes further brain injury due to cerebral hypoxia. In these situations, management strategies are used to reduce ICP with the outcome of restoring cerebral blood flow. It is recommended that a threshold ICP >20mmHg in head injury be reduced, which can be achieved through various methods such as patient sedation, artificial ventilation (to induce hypocapnia), medical therapies or by removal of a part of the skull (182).

Like ICP, ISP relates to perfusion of the anterior pituitary gland, such that perfusion pressure must exceed ISP for the pituitary gland to be perfused to maintain normal
hormone function. Blood perfusion to the normal pituitary gland occurs predominantly through the venous portal system. The superior hypophyseal artery (a branch from the internal carotid artery) supplies the hypothalamus, above the pituitary gland, forming a capillary network that drains the hypothalamus via the portal veins to the anterior pituitary gland. The portal veins supplies nutrient delivery to the anterior pituitary gland as well as hormonal signals from the hypothalamus via a complex capillary network (183). The anterior pituitary gland does also receive direct arterial supply from branches off the superior and inferior hypophyseal arteries with differing configurations in individuals (183).

In normal circumstances ISP is assumed to be equivalent to intracranial pressure, approximately 10mmHg. The measurement of ISP has been achieved historically using lumber puncture (LP) needles primed with saline (studies prior to the year 2000) (184) (185) (186), and more recently by fibreoptic catheter inserted into the pituitary adenoma (113) (187) (188) (189) (190) (191). There is no evidence to suggest that one technique is superior over the another.

Unlike ICP, the measurement of ISP is not utilised clinically, but has been described in the investigational setting of pituitary adenomas. ISP has not been measured in the normal pituitary gland; it would be unethical to undertake this measurement in a normal subject as it requires opening of the sellar floor to access the gland. This is not performed unless the sellar floor is being opened for a surgical removal of an adenoma. The closest estimates to normal ISP have been extrapolated from data taken from microadenomas and in the presence of empty sella syndrome. In non-visible and microadenomas mean ISP was found to range between 7-12mmHg (185) (188) and 11 ± 2mmHg in five cases of empty sella syndrome (185).

Higher ISP is postulated to reduce portal blood flow to the pituitary, leading to impairment of hormonal signalling from the hypothalamus and nutrient delivery to the
normal pituitary and necrosis of viable pituitary cells. One study has examined ISP and pituitary blood flow. In this study authors Kruse et al., examined ISP and pituitary perfusion in 48 cases of various types of pituitary adenomas (35% were non-functioning adenomas) (186). Their aim was to correlate ISP and perfusion and define a critical threshold of pituitary perfusion that could lead to ischemia. The technique of lumber puncture needle with saline was used to measure ISP under mildly hypocapnic conditions (PaO2 30-33.7 mmHg, normal reference range 35-45), and pituitary perfusion measured by the washout of Xenon133 injected into the adenoma (undertaken in 14 cases). In this study the median ISP was reported to be 30mmHg (range 6-62). In three cases, ISP was extremely high (40-60mmHg) due to acute apoplexy during the procedure (accidental and iatrogenic), and it was demonstrated that venous perfusion had completely ceased in two cases and markedly reduced in the third case due to the sudden increase in ISP. The authors concluded that as ISP in these adenomas exceeds that of ICP blood supply to the pituitary cannot be solely due to venous supply, but also due to arterial supply (186). As pituitary adenomas grow slowly, this presumably enables time for formation of additional arterial supply, or else there would be infarction as seen in apoplexy.

1.13.1 Intracellular pressure in pituitary adenomas

There have been 10 studies which have examined ISP in pituitary adenomas (Table 1.6). The primary aims of the studies differ as they correlate ISP to different clinical or biochemical aspects of pituitary disease: these include...

1) ISP and adenoma size;
2) ISP and adenoma type;
3) ISP and pituitary hormones;
4) ISP and headache;
5) ISP and apoplexy;
In this literature review, I will focus on the findings of ISP as it related to adenoma size, adenoma type, pituitary hormones and apoplexy as this is of relevance to this thesis.

**Intrasellar pressure and adenoma size**

Five studies have reported on the relationship between ISP and adenoma size. In only two of these studies determining the relationship between was ISP and adenoma size was a principal study aim (185) (188). Three studies have reported ISP and adenoma size as an additional outcome in these studies (113) (189) (191). Most data suggest that ISP in microadenomas is not particularly raised, with only one study not supporting this finding. Authors Lees et al., using the LP technique, found that in five cases of empty sella syndrome mean ISP was lower at 11mmHg, concluding this must be equivalent to intracranial pressure (185). In seven cases of microadenomas (0-5mm in maximal diameter), mean ISP was 12mmHg, and the authors make note of one case with high ISP of 26mmHg, however they express they were unable to explain this aberrant finding. In microadenomas 6-10mm in size, mean ISP was slightly higher at 17mmHg (185). In another study by authors Gondim et al., the mean ISP in 12 microadenomas was 19.2mmHg (188). Contrary to these findings, eight cases of microadenomas examined by Arafah et al., found ISP was elevated with mean of 25.4 ± 6.7mmHg (113). There were no obvious differences in methodology in these above studies to account for the different findings apart from small sample sizes of each study.

The studies that have examined ISP in macroadenomas have used various ways to describe adenoma size; these differences are listed below...

1) Microadenoma (<1cm) and macroadenoma (≥1cm) (113) (185) (188);
2) Binomial (above and below 26mm in maximal diameter) (191);
3) Position: intrasellar, suprasellar, parasellar, sphenoid sinus (185) (191) or (Hardy-Vezina Grade) (188);
4) Volume (188) (189).

Authors Arafah et al. measured size by binomial method of microadenoma versus macroadenoma. Their series only had 8/49 cases of microadenoma but found no difference in mean ISP compared to the macroadenomas (25.4 ± 6.7mmHg vs 28 ± 14mmHg) and concluded ISP was not correlated with size (113).

In a recent study with the largest cohort of 108 cases, authors Hayashi et al., report in their series consisting predominantly non-functioning adenomas (75.7%), higher ISP in smaller macroadenomas <26mm (24.6 ± 12.1mmHg), compared to those ≥ 26mm (18.3 ± 5.8mmHg) (191). Lower ISP in the larger adenomas is likely explained by the larger lesions being invasive, as the authors found that adenomas that had cavernous sinus invasion had lower pressure than those without (17.8 ± 5.8mmHg vs 22.7 ± 11.5mmHg respectively) (191). Similar findings to this were also found by authors Gondim et al., who examined 60 cases of mixed adenoma types, also describing these by position using the grading system of Hardy and Vezina (188). Highest ISP was found in the macroadenomas confined to the pituitary sella (median 32.6mmHg) with lowest ISP occurring in invasive adenomas (median 12.9mmHg).

In contrast to the above two studies, Lees et al., in 80 cases of mixed type macroadenomas, found highest ISP readings in parasellar adenomas with mean ISP of 30mmHg, although similar to other studies, intrasellar macroadenomas had a mean ISP of 22mmHg and those invasive into sphenoid sinus or having perforated with diaphragma sellae above had lower mean ISP of 13mmHg and 12mmHg, respectively (185).
Lastly, two studies have correlated ISP and volume, both finding that there was no relationship between these two variables. Authors Gondim et al., reported largest volume in invasive adenomas, yet ISP was not highest in these lesions, thus concluding that ISP was not correlated to adenoma size (188). Authors Periero-Neto et al., in their study of 25 cases of pituitary adenomas also did not find ISP to be correlated to adenoma volume (189).

In summary, studies are in agreement that with perforation of the diaphragma sellae (above) or sphenoid sinus (below), ISP tends to be lower. This suggests that when adenoma has breached the diaphragma sellae or exposed outside the pituitary floor into the sphenoid sinus, the adenoma is in equilibrium with intracranial pressure or sphenoid sinus pressure respectively. Most studies have found ISP to be raised in intrasellar adenomas although there is no correlation to adenoma size.

_Intrasellar pressure and adenoma type_

In the published studies of ISP, the majority (9/10) studies have reported ISP in mixed types of pituitary adenomas with non-functioning adenomas contributing 29-75% of the cohorts. Two studies have reported ISP readings specifically in cases of NFPMAs in their series but with different ISP results. In the first study, mean ISP in 8 cases of NFPMA was 28.6 ± 12.6mmHg and in 16 cases with functioning adenomas mean ISP was 19.1 ± 11mmHg: the difference between the mean ISP values was not statistically significant (p=0.18) (184). In the second study, mean ISP in 18 cases of NFPMA was described as not being different to 42 cases with functioning adenomas, with reported mean ISP of 18.7 ± 10.8mmHg (188).
Intrasellar pressure and hormone function

There have only been two studies that have examined the relationship between ISP and hormone function in mixed type of pituitary adenomas. Both studies found that in those without any pre-operative hormone deficiencies ISP was significantly lower with mean of 17mmHg and 19mmHg, respectively in each study, compared to those with one or more hormones deficiencies with mean ISP of 23mmHg (p<0.05) (185) and 33.6mmHg (<0.005) (113), respectively in each study. In the first study by authors Lees et al., ISP was also found to be low in those with a large number (4-5) of hormone deficiencies (185). A plausible explanation for this later finding may be that these cases had very large and invasive adenomas.

Only one study has examined ISP and post-operative hormone status. Hormone “recovery” was the outcome of interest, but the details of the individual hormone axes were not given (113). Although not reaching statistical significance (P=0.075), it was found in 49 cases studied that those with higher ISP (37 ± 13.6mmHg) were actually more likely to recover a hormonal axis compared to those with lower ISP (28.7 ± 13.2mmHg) (113). No study to date has reported on ISP and its relationship to the number of hormone deficiencies post-operatively (apart from one study of ISP in pituitary apoplexy, and this is discussed below).

Studies of ISP have also examined the relationship of ISP to prolactin levels. As mentioned earlier in this literature review (page 31) prolactin is the one pituitary hormone which is under inhibitory control from dopamine signalling from the hypothalamus. Any disruption to the pituitary stalk that inhibits dopamine signalling leads to a mildly raised prolactin (“stalk effect”); this occurs in 38.5-44.8% of NFPMAs, (20) (112). The presence of a normal or mildly raised prolactin in NFPMAs may demonstrate intact pituitary gland reserve as it has been previously observed that after removal of pituitary adenomas, those with higher prolactin levels pre-surgery had
a greater propensity of restoration of other hormonal axes (106) (109) (110) (113) (192) (193).

Five studies in total have examined ISP and its relationship to prolactin in mixed type of pituitary adenomas, but the results are inconsistent. Four studies have found that higher ISP is positively correlated with prolactin levels at presentation (113) (184) (185) (191), but this relationship was not found by one another group (194).

Intrasellar pressure and apoplexy

One study has examined ISP specifically in those presenting with apoplexy. Authors Zayour et al., found markedly elevated ISP in their 13 cases of apoplexy with mean ISP of 44.1 ± 11.1 mmHg (range 25–58 mmHg) compared to a previous series of NFPMA with apoplexy with mean ISP of 33.6 ± 13.4 mmHg, (p=0.04) (187). The authors also reported on the relationship of ISP to hormonal outcomes in these cases of apoplexy finding that those cases (6/13) that had persistent post-operative hormone deficiency had extremely elevated mean ISP of 55.9 ± 2.4 mmHg compared to the remaining cases who had either maintained normal or recovered hormone function with mean ISP of 35.9 ± 7.3 mmHg, p<0.01 (187). The authors suggested that in cases with extremely elevated ISP at the time of apoplexy, raised ISP results in ischaemic necrosis of the anterior pituitary gland, and thus less propensity for hormone recovery. This was further exemplified by their finding of an inverse relationship seen between ISP and prolactin levels: the cases with persistent hypopituitarism post-operatively (and highest ISP) had markedly lower prolactin levels compared to the remaining seven cases who had either maintained or recovered hormone function that had normal or elevated prolactin. Thus, in contrary to the situation of stable (non-apoplectic) pituitary adenomas, ISP appears to be extremely elevated in pituitary apoplexy; additionally, there is an inverse relationship with prolactin levels, and less propensity for hormone recovery of hormone axis in those with extremely high ISP. These findings
relate to this small cohort studied and would need to be confirmed by a larger study of cases with apoplexy.

In summary, the principal finding in the literature of ISP in pituitary adenomas suggests that there is no relationship found between ISP and adenoma size but there is limited data that has found higher ISP to be associated with hormone deficiencies at presentation. This later finding makes biological sense as higher ISP is known to compromise perfusion to the pituitary gland. The situation of extremely high ISP occurring in apoplexy exemplifies even further the pathological role of raised ISP in compromising hormone function. There appears to be relationship between ISP and prolactin levels, but this has not been confirmed by all studies. Overall however, the influence of ISP to hormone function, especially in NFPMAs, which have the highest prevalence of hormone deficiencies, has not been extensively explored and understood. By clarifying the pathophysiology of how ISP influences hormonal status in NFPMAs may translate to improving clinical outcomes in this patient group.

In the final chapter of this thesis (Chapter 5), I seek to explore ISP in a cohort with NFPMAs and determine its relationship to adenoma size and hormonal outcomes, in order to further understand the mechanisms of hormone deficiencies in NFPMAs.
Table 1.6 Literature review of studies investigating intrasellar pressure (ISP)

<table>
<thead>
<tr>
<th>Study, Author, Year, Location</th>
<th>Cases</th>
<th>Aims</th>
<th>Technique used</th>
<th>Adenoma types</th>
<th>Mean ISP mmHg (SD) Range</th>
<th>Findings</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lees, 1987, Uk (184)</td>
<td>24</td>
<td>Explore ISP related to prolactin and stalk compression</td>
<td>LP needle with Saline normotension, normocapnia</td>
<td>Multiple 29% NFA</td>
<td>23(2.5) (2-51) NFA 31.8 (4) No suprasellar extension 20 (3) Suprasellar 27 (3)</td>
<td>Higher ISP associated with higher prolactin</td>
</tr>
<tr>
<td>Kruse, 1992, Netherlands (186)</td>
<td>48</td>
<td>Correlate ISP to pituitary blood flow</td>
<td>LP needle with Saline hypocapnia (PaC0₂ 30-33.7mmHg)</td>
<td>Multiple 35% NFA</td>
<td>Median 30 Range (8-62)</td>
<td>Extremely high ISP (40-50) can cease venous perfusion to pituitary. Shown in 3 acute apoplexy cases. Increasing ICP can also increase ISP (seen by jugular venous compression) No difference in ISP between adenoma type</td>
</tr>
<tr>
<td>Lees, 1994, UK (185)</td>
<td>107</td>
<td>Correlate ISP to adenoma size and hormone function</td>
<td>Blunt needle into adenoma (“refined technique”) Saline injection only if needed. normotension, normocapnia</td>
<td>Multiple characterised by size criteria and invasion</td>
<td>0-5mm adenoma 12 (3) 6-10mm adenoma 17 (2) Intrasellar (&gt;10mm) 22 (2) Suprasellar 21 (2) Parasellar 30 (2) Sphenoid sinus 12 (1) Empty Sella 11 (2)</td>
<td>Highest ISP in parasellar adenomas</td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Pre-operative hormones:</td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Higher ISP if have 1-3 hormone axis deficient But not if 4-5 hormone deficiencies (same ISP as non-hypopituitary cases)</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Higher ISP if raised prolactin (Stalk compression syndrome)</td>
</tr>
<tr>
<td>Kruse, 1995, Netherlands (194)</td>
<td>42</td>
<td>Correlate ISP to prolactin</td>
<td>LP needle with Saline hypocapnia (PaC0₂ 30-33.7mmHg)</td>
<td>Multiple 36% NFA</td>
<td>ISP if raised prolactin 28 (4) (18-43) vs normal prolactin 30 (2) (8-58)</td>
<td>No difference in ISP between those with raised or normal prolactin (p=0.69)</td>
</tr>
</tbody>
</table>

NFA: non-functioning adenomas
<table>
<thead>
<tr>
<th>Study, Author, Year, Location</th>
<th>Cases</th>
<th>Aims</th>
<th>Technique used</th>
<th>Adenoma types</th>
<th>Mean ISP mmHg (SD) Range</th>
<th>Findings</th>
</tr>
</thead>
<tbody>
<tr>
<td>Arafah, 2000, US (113)</td>
<td>49</td>
<td>Correlate ISP to headaches, prolactin &amp; hormone function</td>
<td>Fibre-optic catheter, Stable wave form 30-60 seconds</td>
<td>Multiple 85% macroadenoma (&gt;10mm)</td>
<td>28.8 (13.5) (7-56) Micro 25.4 (6.7) (18-35) Macro 28 (14) (7-56)</td>
<td>Higher ISP in those with headaches, raised prolactin and hormone deficiencies at presentation Higher ISP in those with hormone recovery post-surgery, but not significant 37(13.6) vs 28(13.2), p=0.075 ISP not correlated with adenoma size (but small number of micros n=8)</td>
</tr>
<tr>
<td>Zayour, Arafah, 2004 US (187)</td>
<td>13</td>
<td>Correlate ISP to apoplexy and hormone function</td>
<td>Fibre-optic catheter, Stable wave form 30-60 seconds</td>
<td>Apoplexy &lt;1 week duration</td>
<td>44.1 (11.1) (25-58) Other macroadenomas without apoplexy 33.6 (13.4)</td>
<td>Higher ISP in apoplexy compared to no apoplexy macroadenomas (p=0.04) Higher ISP in those with lower prolactin (p&lt;0.01) ISP not related to adenoma size (p=0.34) Less hormone recovery seen in those with highest ISP and lowest prolactin</td>
</tr>
<tr>
<td>Gondim, 2006 Brazil, (188)</td>
<td>60</td>
<td>Correlate ISP to adenoma volume</td>
<td>Fibre-optic catheter Stable wave form 60 seconds. normotension, normocapnia</td>
<td>Multiple 30% NFA</td>
<td>18.7 (10.8) (2-51)</td>
<td>ISP highest in intrasellar macroadenomas median 32.6 (Hardy Vezina II) Lowest ISP no visible adenoma median 7.0mmHg No difference ISP in types of adenoma</td>
</tr>
<tr>
<td>Periero-Neto, 2010 Brazil (189)</td>
<td>25</td>
<td>Correlate ISP to headache severity, visual field abnormality</td>
<td>Fibre-optic catheter ISP measured for 10 minutes. Mean of 30 readings including 3 Valsalva manoeuvre</td>
<td>Multiple 64% NFA</td>
<td>33.3 (21.1) (13.9-67.1)</td>
<td>ISP not correlated with adenoma volume, headache severity, visual field abnormality or QOL</td>
</tr>
<tr>
<td>Hayashi, 2015 Japan (190)</td>
<td>7 cases 12 control</td>
<td>Correlate ISP in those with severe headache compared to those without headache</td>
<td>Fibre-optic catheter Valsalva manoeuvre before and after sellar floor removed</td>
<td>NFA</td>
<td>41.5 (8) (34-59) with headache vs 22.2 (10.6) (16-30) without headache</td>
<td>ISP was raised in seven cases with severe cases with headaches compared to 12 cases without headaches</td>
</tr>
</tbody>
</table>

NFA: non-functioning adenomas
<table>
<thead>
<tr>
<th>Study, Author, Year, Location</th>
<th>Cases</th>
<th>Aims</th>
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<th>Mean ISP mmHg (SD) Range</th>
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</tr>
</thead>
<tbody>
<tr>
<td>Hayashi 2018 Japan (191)</td>
<td>108</td>
<td>Correlate ISP in those with severe headache compared to those without headache</td>
<td>Fibre-optic catheter. Final pressure after settling</td>
<td>Multiple 75.7% NFA</td>
<td>35.6 (9.2) with headache, 15.8 (5.2) without headache</td>
<td>ISP highest in those headaches, ISP higher in those with raised prolactin, ISP higher in intrasellar adenomas, lower in suprasellar and cavernous sinus invasion, ISP higher in adenomas with cysts and haematoma</td>
</tr>
</tbody>
</table>

NFA: non-functioning adenomas
1.14 The scope of this thesis

This thesis aims to explore clinical aspects of the two most prevalent forms of pituitary adenomas, prolactinomas and non-functioning adenoma (in particular non-functioning pituitary macroadenomas undergoing surgery), that are of clinical importance to the daily management of patients with these conditions. These clinical aspects are:

1) Prolactinomas and the risk of cabergoline-associated valvulopathy (CAV);
2) Non-functioning pituitary macroadenomas (NFPMAs) and...
   a. Surgical outcomes of regrowth and recurrence during long-term follow-up;
   b. Hormonal outcomes of operated cases;
   c. Intrasellar pressure (ISP) and its relationship to adenoma size and hormonal outcomes.

1.14.1 Chapter 2: The risk of cabergoline-associated valvulopathy in prolactinoma patients

As discussed in Chapter 1 of the literature review, medical therapy with dopamine agonists (DAs), particularly cabergoline, is primary therapy for prolactinoma as it is highly efficacious at reducing prolactin levels and reducing adenoma size (28). But in January 2007, two studies reported an association between cabergoline use and an increased risk of valvular heart disease in patients with Parkinson’s disease (PD) (42) (43). Supported by other studies in PD this led to the United States food and drug administration (FDA) and product disclosure statement to issue recommendations in July 2011 for regular echocardiograms every 6-12 months for all patients prescribed cabergoline. Understandably, this concerned endocrinologists as cabergoline is a widely prescribed treatment for prolactinomas.

Until 2019, there had been no guidelines for endocrinologists to follow on how to best screen for CAV except a “remark” in the 2011 Endocrine Society guidelines for the management of hyperprolactinaemia, that echocardiograms may be required for those on
high dose therapy, but those taking smaller doses (<2mg/week), likely will not require regular echocardiography (28). This remark was based on six case control studies of 500 study subjects at that time. These individual studies showed variable results with some showing an increased risk of valvular disease, but it was unclear if it was specifically cabergoline-associated valvulopathy (CAV). In 2012 when I commenced this study, there were no Australian data regarding the risk of CAV in prolactinoma patients. Thus, the aims of Chapter 2 Publication 1 are to:

1) Clarify the definition of cabergoline-associated valvular heart disease (CAV);
2) Examine the risk of CAV in prolactinoma patients in a local Australian cohort;
3) Undertake a systematic review of the literature to determine the prevalence of CAV;
4) Propose a method for screening for CAV in prolactinoma patients.

The relevance of this study was to clarify the definition of CAV (as this was not clear in the literature) and assist in forming recommendation as to how this patient group be optimally screened for CAV.

Chapter 2 Publication 2

From this publication above, it was identified by systematic review and by clarifying the definition of CAV, that only two cases of CAV have occurred world-wide. The findings of this study lead to the recommendation of screening for CAV should be undertaken by a simple annual cardiovascular examination rather than by serial echocardiograms. Through this approach, a third case of CAV was discovered at the local centre. Chapter 2, Publication 2 describes the details of this case, and supports the conclusion of the value of cardiovascular examination as an optimal screening method for CAV.
Chapter 2 Publication 3

Publication 3 is a “Letter to the Editor” I wrote in in 2019. This was in response to a meta-analysis examining the risk of valvular heart disease in prolactinoma patients taking cabergoline, that concluded that there was an increased risk of tricuspid regurgitation (54). I wrote this letter as I did not agree with the study conclusions based on the knowledge that I have acquired though studying this subject area.

1.14.2 Chapter 3: The risk of regrowth and recurrence in operated cases of non-functioning pituitary macroadenomas at a single Australian centre

Trans-sphenoidal surgery is the treatment of choice for NFPMAs which exhibit symptoms of mass effect including visual disturbance, headache and impaired pituitary hormone function. The long-term outcomes of NFPMAs following surgery has been considered unclear, with varying rates of regrowth and recurrence from data across the world. However, data are consistent that the presence of residual disease is a strong risk factor for regrowth. What remains poorly defined are other risk factors for regrowth and recurrence, optimum management of residual disease and optimal duration of long-term radiological surveillance (82) (195). At the time of undertaking this study there were no Australian data regarding the surgical outcomes of NFPMAs. Thus, the aims of Chapter 3 are to:

1) Describe surgical outcomes and regrowth rate of NFPMAs in a local cohort;
2) Identify risk factors that may predict regrowth and recurrence, in order to assist with planning of long-term radiological follow-up.

The relevance of this study is to understand the local Australian experience at a centre which specialises in pituitary care to ensure the outcomes are equal to that of the international experience. Furthermore, to discover any risk factors for regrowth and recurrence that would be of clinical importance and assist in rationalising follow-up.
1.14.3 Chapter 4: Hormonal outcomes in operated cases of non-functioning pituitary macroadenomas at a single Australian centre

Hormone deficiencies are a well-known morbidity of NFPMAs and the presence of one or more hormonal axis deficiency necessitates lifelong follow-up with endocrinologists. Many series have reported on hormone deficiencies in operated cases of NFPMAs, however there have not been any Australian data published. Furthermore, there has been limited information about gender differences in hormonal outcomes. The aims of Chapter 4 of this thesis are to:

1) Describe the hormonal status at presentation and post-operatively in a local Australian cohort with NFPMAs;
2) Explore gender differences in hormone axis deficiency at presentation and post-operatively.

The relevance of this study is to understand the local Australian experience at a centre which specialises in pituitary care. The understanding of hormonal outcomes is vital to the management and counselling of patients prior to surgery.

The novel findings of this study have led to the development of following study in Chapter 5, which explores the role that raised intrasellar pressure (ISP) has in leading to hormonal deficiencies.

1.14.4 Chapter 5: The relationship between intrasellar pressure, adenoma size and hormonal outcomes in operated cases of non-functioning pituitary macroadenomas

Intrasellar pressure (ISP) is the pressure within the pituitary gland as it sits within the pituitary fossa. ISP is assumed to be equivalent to pressure within the brain (10mmHg) and data has shown that ISP is elevated in pituitary adenomas. A small number of studies have examined ISP in mixed types of pituitary adenoma; this has been described in the literature
review in Chapter 1. In small or non-visible adenomas data on ISP varied with some studies showing low pressure but other studies reporting high pressure similar to macroadenomas. Studies, however, have not shown that ISP is related to adenoma size. Two studies have found that hormone deficiencies at presentation may be related to higher ISP (113) (185).

The novel findings from the Chapter 4, of the favourable hormonal outcomes of pre-menopausal females, led to the hypothesis that smaller adenomas (which are known to occur in pre-menopausal females) may be associated with lower ISP. I wanted to further understand what ISP is and its relationships to adenoma size and hormonal outcomes in operated cases of NFPMAs. Thus, the principal aims of Chapter 5 are:

1) To determine the relationship of ISP to adenoma size in NFPMAs;
2) To determine the relationship between ISP and pre- and post-operative hormonal outcomes in NFPMAs.

1.14.5 Overall Thesis Framework

This thesis is composed of a series of self-contained chapters addressing the aims listed above. Each chapter begins with an introduction, followed by a published manuscript and ending with a short conclusion. Chapter 5 is unpublished material, not submitted for publication, so is presented in a traditional chapter format. This thesis is linked together by a summary of the key findings (Chapter 6) and conclusion and future directions (Chapter 7). References are presented at the end of this thesis followed by three appendices.
Chapter 2: The risk of cabergoline-associated valvulopathy in prolactinoma patients

2.1 Introduction

Prolactinomas are the commonest type of pituitary adenoma with a prevalence of 35-53 per 100,000 population. Prolactinomas are commonly encountered and managed by general practitioners, endocrinologists and fertility specialists. Whether these are microadenomas or macroadenomas first line therapy is with dopamine agonists (DAs), of which cabergoline is the most widely prescribed. Cabergoline has been associated with valvular heart disease in patients with Parkinson’s disease (PD), so understandably this concern extended to those with prolactinomas. The consequences of valvular heart disease can include cardiac failure and possibly the requirement of cardiac valve replacement. Since 2007, literature emerged from prolactinoma cases exploring the prevalence of cabergoline-associated valvular heart disease (CAV).

Publication 1 comprises of a clinical study aimed to detect the prevalence of CAV in the local cohort, includes a systematic review which was undertaken to clarify the prevalence of CAV in the literature and examines optimal screening methods for CAV.

Publication 2 is a case report of CAV detected at this centre and discovered following the screening recommendation of the study above. This case report also highlights more recent literature published on the topic since the systematic review presented above.

Publication 3 is a “Letter to the Editor” in response to a 2019 meta-analysis published which concluded that cabergoline is associated with a risk of tricuspid regurgitation. This letter highlights the salient issues uncovered by this thesis that have been able to contribute to a robust discussion about CAV.
2.2 Publication 1

The need for annual echocardiography to detect cabergoline-associated valvulopathy in patients with prolactinoma: a systematic review and additional clinical data.

Caputo C, Prior D, Inder W.

Lancet Diabetes Endocrinology. 2015 Nov; 3(11); 906-13
The need for annual echocardiography to detect cabergoline-associated valvulopathy in patients with prolactinoma: a systematic review and additional clinical data

Carolea Capote, David Price, Warrick Hinder

Present recommendations by the US Food and Drug Administration advise that patients with prolactinoma treated with cabergoline should have an annual echocardiogram to screen for valvular heart disease. Here, we present new clinical data and a systematic review of the scientific literature showing that the prevalence of cabergoline-associated valvulopathy is very low. We prospectively assessed 40 patients with prolactinoma taking cabergoline. Cardiovascular examination before echocardiography detected an audible systolic murmur in 10% of cases (all were functional murmurs), and no clinically significant valvular lesion was shown on echocardiogram in the 90% of patients without a murmur. Our systematic review identified 21 studies that assessed the presence of valvular abnormalities in patients with prolactinoma treated with cabergoline. Including our new clinical data, only two (0.11%) of 1811 patients were confirmed to have cabergoline-associated valvulopathy (three [0.17%] if possible cases were included). The probability of clinically significant valvular heart disease is low in the absence of a murmur. On the basis of these findings, we challenge the present recommendations to do routine echocardiography in all patients taking cabergoline for prolactinoma every 12 months. We propose that such patients should be screened by a clinical cardiovascular examination and that echocardiograms should be reserved for those patients with an audible murmur, those treated for more than 5 years at a dose of more than 3 mg per week, or those who maintain cabergoline treatment after the age of 50 years.

Introduction

Dopamine agonists and in particular cabergoline are the treatment of choice for patients with prolactinoma. In 2007, findings of two studies showed the association between cabergoline and an increased risk of valvular heart disease in patients with Parkinson's disease, in whom dopamine agonists are a major class of therapeutic agent used for symptomatic management in Parkinson's disease. Along with findings of other studies of Parkinson's disease, these results led to the US Food and Drug Administration (FDA) issuing recommendations in July, 2011, for regular echocardiograms every 6–12 months in all patients taking cabergoline. No guidelines are available for endocrinologists except a remark in the 2011 Endocrine Society guidelines for the management of hyperprolactinemia, which state that echocardiograms might be necessary for individuals on high-dose cabergoline treatment, but those on low doses (<2 mg per week) will probably not need regular echocardiography. This remark was based on six case-control studies of 508 study participants available at the time the guidelines were written. A recent clinical review summarising the scientific literature found no conclusive evidence that cabergoline causes clinically significant valvular heart disease, but called for evidence-based criteria to identify patients at risk and the creation of appropriate screening protocols. The authors of the Review also suggested minimising exposure to cabergoline, for example by considering surgical resection of the pituitary adenoma or careful withdrawal of the drug in responders.

The mechanism of valvular pathology as a cabergoline-induced adverse event is biologically plausible; the drug stimulates serotonin receptor subtype 5-HT₆, which is expressed in heart valves and mediates mitogenesis and proliferation of fibroblasts. However, cumulative evidence shows a very low prevalence of cabergoline-associated valvulopathy in patients with prolactinoma. Most studies have focused on prevalence of any valvular lesion as detected by echocardiogram, with little reference to distinguishing cabergoline-associated valvulopathy (defined as the triad of moderate or severe regurgitation, associated with a restricted and thickened valve) from other frequently recorded valvular abnormalities in patients with prolactinoma. Echocardiograms have limitations; their interpretation needs to incorporate clinical history and findings of the cardiovascular examination. This finding suggests that the prevalence of clinically significant valvular disease is low when a normal examination is recorded. Importantly, examination by non-cardiologists distinguishes well between innocent murmurs and valvular heart disease. In this Review, we present new clinical data and results of a systematic review to estimate the prevalence of cabergoline-associated valvulopathy in patients treated with cabergoline and whether routine annual echocardiograms in such patients are justified. Our clinical data are from a cohort of 40 patients with prolactinoma taking cabergoline, in whom cardiovascular examination was used as a screening method for valvular...
heart disease, followed by transthoracic echocardiography, to assess whether cardiovascular examination alone is sufficient to exclude significant valvular pathology in routine clinical practice. We also make recommendations for the use of echocardiography in this clinical context.

Prospective assessment of cardiovascular examination versus echocardiography in patients with prolactinoma

Between June 1, 2009, and March 6, 2012, we prospectively recruited 49 consecutive patients at St Vincent’s Hospital Melbourne, Australia, a tertiary institution with a dedicated multidisciplinary pituitary clinic, and from private practices of authors CC and WJ (panel 1). Nine patients were excluded for not attending echocardiogram, one of whom (a 70-year-old man) died during follow-up.

Table 1 shows demographic features of the 40 patients who completed echocardiography. 19 (48%) were women; the women in our cohort were significantly younger than the men (mean 39-6 [SD 12-2] vs 48-9 [12-1] years, p=0.03). Men had a higher prevalence of macroprolactinoma than women (20 [95%] of 21 men vs eight [42%] of 19 women, p=0.001). Mean cumulative dose of cabergoline was 391.3 mg (SD 532.7) during treatment for 21-2 months (SD 66-2). Weekly and total cumulative doses did not differ between sexes. Six patients had previous exposure to bromocriptine. The cohort was otherwise healthy with few cardiovascular risk factors, and only one patient had cardiorespiratory symptoms with shortness of breath on exercise.

Clinical cardiovascular examination showed cardiac murmurs in four (10%) of 40 patients (three women aged 23, 39, and 62 years, and one man aged 52 years) and were all soft (grade 2/6) ejection systolic murmurs associated with normal first and second heart sounds, suggestive of benign or innocent murmurs.1 No patient had clinical signs of left or right ventricular failure or pulmonary hypertension.

On the basis of transthoracic echocardiography, we found no evidence of moderate or severe valvular lesions or cabergoline-associated valvulopathy (table 2). The four patients with cardiac murmurs on auscultation had normal valve morphology. We noted five cases (12%) of valvular abnormality detected by valvular thickening: two cases of myxomatous mitral valve disease and three cases of age-related aortic sclerosis. The two cases of myxomatous mitral valve disease had mild mitral regurgitation with normal left ventricular systolic function. Of the three patients with age-related aortic sclerosis, one had no regurgitation, and one had trivial regurgitation with normal valve function. The third patient had mild aortic regurgitation. These five cases with valvular abnormalities had normal cardiovascular examination. These results suggest a low prevalence of cabergoline-associated valvulopathy in a cohort with a moderate cumulative dose of cabergoline. Cardiac examination by non-cardiologists correctly excluded clinically significant valvular pathology and correctly identified benign murmurs. Interpretation of valvular lesions detected by echocardiography needs careful assessment, in terms of the nature and cause of the lesion because the presence of mild regurgitation itself does not equate to cabergoline-associated valvulopathy.

Systematic review

Methods

We did a literature review with the aim of summarising the published scientific literature to estimate the prevalence of cabergoline-associated valvulopathy in patients with prolactinoma. We searched PubMed and Embase from Aug 1, 1960, to July 31, 2014, with the search terms “prolactinoma and cabergoline” and “cabergoline and cardiac” to identify original studies and cases of cabergoline-associated valvulopathy in patients with prolactinoma. We included original research studies published in English (figure). We estimated prevalence with confirmed cases of cabergoline-associated valvulopathy, and with possible but unconfirmed cases. The total of all patients given cabergoline in studies and case reports identified, including data from our own cohort reported in this Review, was used as the denominator to estimate prevalence. We worked out mean age, follow-up

Panel 1: Methods for prospective assessment of cardiovascular examination and echocardiography in patients with prolactinoma treated with cabergoline

Patient characteristics

Patients were included if they had received cabergoline for at least 12 months for the treatment of prolactinoma and excluded if they had known ischaemic cardiac disease, coronary syndrome, atrioventricular block, atrial fibrillation, or previous use of other ergot-derived drugs implicated in fibrotic valvulopathy. Written informed consent was obtained from all patients, and the study was done with approval from the human research ethics committee at St Vincent’s Hospital Melbourne, Victoria, Australia.

Cardiovascular examination

Clinical cardiovascular examinations were done during routine office review by one of two consultant endocrinologists (CC or WJ). Parameters of manual blood pressure, pulse rate, and percutaneous occlusion were done in a medically accepted standard of examination. Echocardiograms were done with GE Vivid 7 (GE Medical, Horten, Norway) and reported by a cardiologist (DP), who was masked to clinical cardiovascular findings, but was aware of cabergoline exposure. Quantitative measurements for regurgitant valves were made according to recommendations of the American Society of Echocardiography, with detailed reporting of valvular morphology.12 Cabergoline-associated valvulopathy was defined as the triad of moderate to severe regurgitation, associated with a restricted and thickened valve. This definition derived by clinical evaluation of a valvular lesion being that of at least moderate severity, the echocardiographic findings from the confirmed case of cabergoline-associated valvulopathy described by Ganwood and colleagues13 and by what has been previously described as other findings of ergot-associated valvulopathy.12 Calculation is not a feature of cabergoline-associated valvulopathy.

Statistical analysis

We present demographic information as percentages and means with SD. Data were compared by sex with χ² for categorical data and mean comparison tests for continuous variables. We used STATA (version 12.1) for analyses.
Valvulopathy unlikely. The same group published another series of 40 patients who had baseline (ie, at the start of cabergoline treatment) and follow-up echocardiograms at 5 years, with no new valvular lesions detected. In this study, no cases of moderate tricuspid regurgitation were reported.

Two studies reported one case each of cabergoline-associated valvulopathy that we deem as definite. One case report of fulminant cardiac failure secondary to severe tricuspid regurgitation was reported in a man aged 59 years with prolactinoma taking cabergoline with a cumulative dose of 252 mg during 3-5 years. This patient presented with symptoms (evidence of left ventricular failure), and echocardiography findings showed moderate tricuspid regurgitation and severe mitral regurgitation associated with thickening and restriction but without calcification of the mitral valve. Histology of the valve postoperatively confirmed cabergoline-associated valvulopathy (and confirmed the absence of calcification). Another case of cabergoline-associated valvulopathy was reported by Gu and colleagues, who compared the effects of masked versus unmasked reporting of echocardiograms. The patient identified in this study had moderate mitral regurgitation that the investigators concluded was due to cabergoline-associated valvulopathy.

We judge one case reported in the literature as possible cabergoline-associated valvulopathy. Bhat and colleagues studied a 60-year-old woman with right heart failure and moderate tricuspid regurgitation who had taken a cumulative dose of 48 mg cabergoline for 48 months. The patient presented symptomatically with dyspnoea, ankle swelling, and abdominal discomfort, and clinical examination found a raised jugular venous pressure, systolic murmur, tender hepatomegaly, and pitting oedema. However, the investigators reported no morphological description of the tricuspid valve to confirm cabergoline-associated valvulopathy.

In a large cross-sectional study by Drake and colleagues, of 711 participants given cabergoline and 32 given bromocriptine, moderate regurgitation was recorded in 23 participants. No severe valvular lesions were detected. The researchers reported two cases of thickened valves and eight cases of valve restriction, but whether these findings arose in the same patients with regurgitation is unclear. The investigators did not confirm the presence of cabergoline-associated valvulopathy. So far, including the clinical data presented here, 1811 individuals with prolactinoma taking cabergoline have been studied with detailed echocardiograms. Collectively, the results of these studies show a very low prevalence of cabergoline-associated valvulopathy as defined by the triad of moderate or severe regurgitation associated with a restricted and thickened valve. If two confirmed cases by Cowood and colleagues and Gu and colleagues are included, we estimate that the prevalence of cabergoline-associated valvulopathy is 0.11% (2/1811). At most,

duration, and cumulative doses from studies reporting these data, and expressed them as mean (SD).

Valvulopathy in patients with prolactinoma

Our literature search identified 21 studies, all of which were included: 13 case-control studies, two retrospective studies, two prospective follow-up studies, a study comparing echocardiograms being read either masked or unmasked to cabergoline treatment, a cross-sectional study of 711 patients, and two individual case reports (table 3).

Table 2: Patient clinical characteristics

<table>
<thead>
<tr>
<th>n</th>
<th>Women</th>
<th>Men</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sex</td>
<td>40</td>
<td>19 (48%)</td>
</tr>
<tr>
<td>Age (years)</td>
<td>44.1 (12.8)</td>
<td>39.0 (12.8)</td>
</tr>
<tr>
<td>Hypertrophic cardiomyopathy</td>
<td>32</td>
<td>16</td>
</tr>
<tr>
<td>Mitral stenosis</td>
<td>28</td>
<td>10</td>
</tr>
<tr>
<td>Iliac gynecomastia</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>Cardiac conduction disorders</td>
<td>3 (6.4%)</td>
<td>3 (6.4%)</td>
</tr>
<tr>
<td>Cardiac conduction disorders (mg)</td>
<td>5 (7.12, 22.7-17.8)</td>
<td>5 (7.12, 22.7-17.8)</td>
</tr>
<tr>
<td>Treatment duration (months)</td>
<td>7.2</td>
<td>12.2-21.5</td>
</tr>
<tr>
<td>Treatment duration (months)</td>
<td>7.2</td>
<td>12.2-21.5</td>
</tr>
<tr>
<td>Treatment duration (months)</td>
<td>7.2</td>
<td>12.2-21.5</td>
</tr>
<tr>
<td>Treatment duration (months)</td>
<td>7.2</td>
<td>12.2-21.5</td>
</tr>
<tr>
<td>Treatment duration (months)</td>
<td>7.2</td>
<td>12.2-21.5</td>
</tr>
</tbody>
</table>

Table 2: Echocardiographic findings from 40 patients with prolactinoma treated with cabergoline.

<table>
<thead>
<tr>
<th>Mitral valve</th>
<th>Aortic</th>
<th>Tricuspid</th>
<th>Pulmonary</th>
</tr>
</thead>
<tbody>
<tr>
<td>No regurgitation</td>
<td>34 (54%)</td>
<td>30 (49%)</td>
<td>25 (38%)</td>
</tr>
<tr>
<td>Severe regurgitation</td>
<td>22 (55%)</td>
<td>7 (2%)</td>
<td>24 (35%)</td>
</tr>
<tr>
<td>Mild regurgitation</td>
<td>2 (5%)</td>
<td>2 (5%)</td>
<td>2 (5%)</td>
</tr>
<tr>
<td>Moderate to severe regurgitation</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Valve thickness</td>
<td>2±1</td>
<td>1±1</td>
<td>0</td>
</tr>
</tbody>
</table>
including the additional possible case reported by Bhat and colleagues, we estimate the prevalence to be 0.17% (3/1811).

Cumulative doses of cabergoline in patients with prolactinoma
Echocardiographic studies of patients with prolactinoma taking cabergoline have been based largely on the original case-control study by Zanettini and colleagues in patients with Parkinson’s disease. In that study, the mean cumulative dose of cabergoline was 405 mg (SD 3208) in patients who had moderate and severe valvular lesions, and the mean age of participants was 61.5 years (SD 9.8). The demographic characteristics of patients with prolactinoma identified in the systematic review differ substantially to the cohort with Parkinson’s disease studied by Zanettini and colleagues in terms of age and weekly and cumulative dose. In studies of patients with prolactinoma treated with cabergoline in which means were available, the mean age was 43.7 years (SD 4.9), the cumulative dose was 335-8 mg (170-9), and the duration of treatment was 59-8 months (20-8). Thus, it would take about 59-4 years of treatment to reach the exposure of more than 4000 mg that was associated with valvular lesions in the study by Zanettini and colleagues. Few patients with prolactinoma (5/50 [10%]) need cabergoline treatment at doses greater than 3 mg per week. Too few cases of cabergoline-associated valvulopathy in patients with prolactinoma exist to establish whether there is a threshold dose-effect of cabergoline, but two studies of prolactinomas have reviewed serial echocardiograms longitudinally for 2 and 5 years, without showing any new cases of cabergoline-associated valvulopathy.

Utility of echocardiography to screen for valvular pathology
Like any diagnostic method in medicine, physicians need to be aware that echocardiograms have their limitations. Additionally, knowledge of patient treatment with cabergoline might lead to over-reporting. Indeed, when echocardiograms from patients with prolactinoma taking cabergoline or bromocriptine were reported twice—one group of reporters were told participants had not been exposed to drug, the second group were informed of drug exposure—investigators noted significantly more cases of trivial valvar regurgitation and valvular thickening when unmasked. In the large cross-sectional study of 711 patients taking cabergoline, 28 centers were included in data collection of unmasked echo-cardiograms with different echocardiographic machines and reporters, thus potentially increasing over-reporting bias. Many variables can affect result acquisition, including machine variables (eg, gain settings and beam angles), inter-machine variability, inter-reporting and intra-reporting variability, and clinical parameters such as weight, heart rate, and blood pressure. Incorrect echocardiogram results can arise; without association with patient symptoms and results of cardiovascular examination, these errors can lead to unnecessary and sometimes detrimental consequences. Additionally, costs need to be considered, not only financial cost of echocardiograms (and repeated studies if abnormalities are found), but also emotional and time costs for patients.

In studies of prolactinoma and echocardiogram up to now, the focus has been on reporting all valvular abnormalities present, rather than distinguishing cabergoline-associated valvulopathy, noting that the prevalence of mild valvular regurgitation on echocardiography in the general population is 20%. Furthermore, no study has associated the echocardiographic findings with detailed clinical cardiovascular examination within a cohort. The relevant outcome is clinically significant disease (moderate or severe regurgitation) that establishes a clinical decision about whether to stop or continue cabergoline. If a valvular lesion is found, a clinician needs to know whether the change is age related, myxomatous, or cabergoline-associated valvulopathy to decide how to continue with treatment. Tricuspid valve tethering area and mitral valve tenting (described in some
studies\(^{1,2,3,4}\) are useful measurements for comparing defined patient groups to quantify the amount of valve restriction, but can be affected by user and machine variability. Therefore, these measurements are not reliably reproducible and are only surrogate markers of subclinical changes in the valve. In clinical practice, these measurements are not markers of echocardiograms that would change medical management.

Utility of cardiovascular examination to screen for valvular pathology
Cardiovascular examination is a sufficiently accurate method to assess clinically significant valvular heart disease when done either by cardiologist or non-cardiologist.\(^{5,6,7}\) Of 34 asymptomatic participants (normal volunteers and patients with connective tissue disorders), a cardiovascular examination had positive and

<table>
<thead>
<tr>
<th>Author, year, reference</th>
<th>Participants, women (%)</th>
<th>Age (years)</th>
<th>Follow-up (months)</th>
<th>Cumulative cabergoline dose (mg)</th>
<th>Masked echocardiograms</th>
<th>Findings</th>
<th>Cabergoline-associated valvularopathy</th>
</tr>
</thead>
<tbody>
<tr>
<td>Case-control</td>
<td>Lucidotti, 2008(^a)</td>
<td>402 subjects, 3 controls (72%)</td>
<td>53 (36)</td>
<td>Median 39 (range 12-218)</td>
<td>Median 264 (range 18-718)</td>
<td>Yes</td>
<td>Two cases of moderate mitral regurgitation, but not significantly different to controls, increased mitral valve testing, mitral valve thickening more than 5% in cabergoline group vs 0% in control group (p=0.08)</td>
</tr>
<tr>
<td>Case-control</td>
<td>Bars, 2008(^b)</td>
<td>42 cabergoline, 31 other dopamine agonists, 36 controls (72%)</td>
<td>Mean 46 (SE 3.8)</td>
<td>Mean 62.4 (SE 4.9)</td>
<td>Mean 68 (SE 9.5)</td>
<td>Yes</td>
<td>No increase in prevalence of moderate or severe mitral regurgitation or tricuspid regurgitation</td>
</tr>
<tr>
<td>Case-control</td>
<td>Walli, 2008(^b)</td>
<td>44 cabergoline, 31 other dopamine agonists, 36 controls (72%)</td>
<td>Mean 48 (11-2)</td>
<td>Mean 44.8 (median 42)</td>
<td>Mean 11 (median 15)</td>
<td>No</td>
<td>No increase in prevalence of mild, moderate, or severe mitral regurgitation in any valve</td>
</tr>
<tr>
<td>Case-control</td>
<td>Gallo, 2008(^b)</td>
<td>39 cabergoline, 26 other dopamine agonists, 36 controls (72%)</td>
<td>36.5 (18-5)</td>
<td>Median 34</td>
<td>415 (90-56)</td>
<td>No</td>
<td>No increase in mild mitral regurgitation, more severe tricuspid regurgitation (24% vs 14%, p=0.001), no changes in valve morphology</td>
</tr>
<tr>
<td>Case-control</td>
<td>Valletta, 2008(^b)</td>
<td>28 cabergoline, 39 controls (52%)</td>
<td>44 (13)</td>
<td>55 (27)</td>
<td>29 (13)</td>
<td>Yes</td>
<td>No difference in moderate regurgitation and associated valve thickening, no cases of severe regurgitation</td>
</tr>
<tr>
<td>Case-control</td>
<td>Heering, 2008(^b)</td>
<td>50 cabergoline, 50 controls (43%)</td>
<td>53.2 (2.2)</td>
<td>Mean 75.2 (41.6)</td>
<td>Mean 44.1 (SE 5.2)</td>
<td>Yes</td>
<td>No cases of moderate- to severe lesions</td>
</tr>
<tr>
<td>Case-control</td>
<td>Tan, 2008(^b)</td>
<td>50 cabergoline, 50 controls (52%)</td>
<td>36 (24-45)</td>
<td>53.9 (39-96)</td>
<td>78 (68-298)</td>
<td>No</td>
<td>No cases of moderate or severe valvular lesions</td>
</tr>
<tr>
<td>Case-control</td>
<td>Nachtigal, 2008(^b)</td>
<td>100 cabergoline, 100 controls (33%)</td>
<td>44 (13)</td>
<td>Mean 49 (SD 4.6)</td>
<td>Mean 25.3 (SD 0.1)</td>
<td>Yes, two cases</td>
<td>No difference in mild or mild to moderate mitral regurgitation, no cases of moderate to severe regurgitation, no cases of moderate to severe lesions</td>
</tr>
<tr>
<td>Case-control</td>
<td>Boguszewski, 2007(^b)</td>
<td>31 cabergoline, 31 bromocriptine, 31 control (29%)</td>
<td>42 (13-3)</td>
<td>37 (23-15)</td>
<td>23 (8-42)</td>
<td>Yes</td>
<td>Increased mitral and aortic valve testing, more mild to moderate regurgitation (73% vs 50%)</td>
</tr>
<tr>
<td>Case-control</td>
<td>Helzner, 2007(^b)</td>
<td>62 cabergoline, 62 bromocriptine, 62 control (29%)</td>
<td>37 (12-4)</td>
<td>42 (3-42)</td>
<td>25 (9-7)</td>
<td>No, three centers</td>
<td>No moderate- to severe lesions, no difference in mild or moderate regurgitation</td>
</tr>
<tr>
<td>Case-control</td>
<td>Benkova, 2007(^b)</td>
<td>30 cabergoline, 30 bromocriptine, 30 control (28%)</td>
<td>38 (6-12)</td>
<td>46 (7-12)</td>
<td>17 (8-12)</td>
<td>Yes</td>
<td>No cases of moderate to severe mitral regurgitation in either group, no difference between groups in clinically relevant regurgitation</td>
</tr>
<tr>
<td>Case-control</td>
<td>Cordba, 2007(^b)</td>
<td>32 cabergoline, 32 control (73%)</td>
<td>38 (10-4)</td>
<td>15 (6-20)</td>
<td>15 (6-20)</td>
<td>Yes</td>
<td>One case of moderate aortic regurgitation in cabergoline group but this was not associated with morphological changes</td>
</tr>
<tr>
<td>Case-control</td>
<td>Lathe, 2007(^b)</td>
<td>34 cabergoline, 34 bromocriptine, 34 control (57%)</td>
<td>58 (14-7)</td>
<td>Median 315 (range 7-515)</td>
<td>Median 277 (range 8-358)</td>
<td>Yes</td>
<td>Nine cases of clinically significant valvular lesion in cabergoline group vs no cases (p=0.02). Eight cases of mild aortic regurgitation, one case of moderate tricuspid regurgitation</td>
</tr>
<tr>
<td>Retrospective,</td>
<td>Desai, 2008(^b)</td>
<td>45 cabergoline (60%)</td>
<td>41 (19)</td>
<td>38 (29)</td>
<td>14 (7)</td>
<td>No</td>
<td>No cases of moderate- or severe valvular lesions</td>
</tr>
<tr>
<td>Retrospective,</td>
<td>Tamarin, 2012(^d)</td>
<td>34 cabergoline, 34 bromocriptine (2%)</td>
<td>45 mean (range 20-67)</td>
<td>6 mean (range 12-128)</td>
<td>15.5 mean (range 9-23)</td>
<td>No</td>
<td>No valve abnormality in any patients</td>
</tr>
</tbody>
</table>

\(^{1,2,3,4}\) \(^{5,6,7}\) 

(continued as next page)
<table>
<thead>
<tr>
<th>Author, year of reference</th>
<th>Participants, women (%)</th>
<th>Age (years)</th>
<th>Follow-up (months)</th>
<th>Cumulative cabergoline dose (mg)</th>
<th>Masked echocardiograms</th>
<th>Findings</th>
<th>Cabergoline-associated valvulopathy</th>
</tr>
</thead>
<tbody>
<tr>
<td>Prospective, 2 year follow-up</td>
<td>Delamere, 2012**</td>
<td>40 cabergoline, 21%</td>
<td>Median 56 (SE 4.4)</td>
<td>Mean 60.4 (SE 4.4)</td>
<td>Yes</td>
<td>Increased aortic valve calcification during 2 years (45.8% in cabergoline group, 29.9% in non-cabergoline group) but no change in thickness of valves, and no new valvular lesions.</td>
<td>No</td>
</tr>
<tr>
<td>Prospective, 1 year follow-up</td>
<td>Anterossa, 2013**</td>
<td>40 cabergoline (21%)</td>
<td>38.7 (21-5)</td>
<td>60</td>
<td>Median 1.4 (range 0.38-12.69)</td>
<td>No mention of moderate to severe tricuspid regurgitation, &gt;10% of patients had mild tricuspid regurgitation.</td>
<td>No</td>
</tr>
<tr>
<td>Case report</td>
<td>Gilla, 2012**</td>
<td>24 cabergoline, 50% of patients noted the effect of mask-valve exposure</td>
<td>49.3 (45)</td>
<td>129.3 (52-8)</td>
<td>Cabergoline dose (40%) [93], both cabergoline and bromocriptine (35%) [96]. Group A investigators told patients were not exposed, group B told cases were taking dopamine agonists.</td>
<td>24/60 moderate to mild mitral regurgitation due to cabergoline-associated valvulopathy, 10 cases of aortic/mitral regurgitation or tricuspid valve and/or aortic valve thickening in group B.</td>
<td>One case</td>
</tr>
<tr>
<td>Cross-sectional</td>
<td>Ditko, 2014**</td>
<td>601 cabergoline alone, 32 bromocriptine, 52 bromocriptine</td>
<td>52 (14-63)</td>
<td>NA</td>
<td>252 (19.3-38)</td>
<td>No, 28 patients</td>
<td>27/60 had mild mitral regurgitation at any valve, no severe lesions. 2 cases of moderate to severe mitral regurgitation in the combined cohort (including bromocriptine group). 11 aortic valve, nine tricuspid valve, three mitral valve. Two patients had aortic valve thickening and one had aortic valve stenosis, none had mitral valve restriction, none had chronic valve restriction, but investigators did not consider presence of the triad of cabergoline-associated valvulopathy.</td>
</tr>
<tr>
<td>Case report</td>
<td>Cazzal, 2009**</td>
<td>1 man taking cabergoline</td>
<td>59</td>
<td>42</td>
<td>252</td>
<td>NA</td>
<td>Severe mitral regurgitation with thickened and restricted valve, with moderate to severe mitral regurgitation in a patient taking cabergoline. Associated valvulopathy of mitral valve postoperatively.</td>
</tr>
<tr>
<td>Case report</td>
<td>Ellis, 2013**</td>
<td>1 woman taking cabergoline</td>
<td>60</td>
<td>48</td>
<td>48</td>
<td>NA</td>
<td>Moderate to severe mitral regurgitation, moderate to severe mitral valve thickening, no mention of this finding in control group. Possible but unlikely.</td>
</tr>
</tbody>
</table>

Data are mean (SEM) or median (IQR) unless otherwise stated.

Table 3: Summary of original research studies examining prevalence of valvular abnormalities in patients with prolactinoma taking cabergoline.

negative predictive values of 92%, and specificity of 98% in the diagnosis of valvular heart disease of mild severity as confirmed by echocardiogram.7 In another study6 of 203 patients in which ascultation done by emergency department physicians had identified a systolic murmur, clinical examination could usually distinguish between a functional (innocent) murmur and clinically significant valvular heart disease. Furthermore, in a study8 of 100 patients referred for echocardiography after discovery of a systolic murmur of unknown cause, only 23% had clinically significant heart disease that was completely missed on clinical examination. A study7 of 227 patients with obesity taking desflurane, an anestrogen associated with valvar heart disease that is proposed to act via the same mechanism as cabergoline, compared ascultation by non-cardiologists with echocardiography. Ausible murmurs were found in 14% of patients compared with in 11% of controls (ie, individuals with obesity but not taking desflurane). In individuals treated with desflurane, the absence of a murmur had a high specificity (89%) and negative predictive value (85%) for excluding mild or worse regurgitation. Patients with abnormalities missed by clinical examination had mild regurgitation and normal valve morphology. The investigators concluded that cardiac ascultation should be the screening method of choice to detect valvular regurgitation in patients taking desflurane because the absence of a murmur predicted the absence of clinically significant valvular pathology.

**Conclusion and recommendations**

In our clinical cohort of patients with prolactinoma treated with cabergoline, cardiovascual examination by the treating endocrinologist detected an audible systolic murmur in 10% of patients, all of which were functional systolic murmurs. The absence of any clinically significant valvular lesion on echocardiogram in the 90% of patients who did not have a murmur is consistent with the scientific literature on the accuracy of physical examination as a screening method. Our study does not have a control
Panel 2: Key messages

- Present recommendations are that people taking cabergoline for treatment of prolactinoma should have an annual echocardiogram to screen for valvular heart disease.
- Findings of studies including 1931 patients with prolactinoma taking cabergoline have not shown an increased risk of clinically significant valvular heart disease.
- The presence of valvular regurgitation is a common finding on echocardiography but does not necessarily signify cabergoline-associated valvulopathy. Morphological features are significant regurgitation associated with a thickened and restricted valve.
- A cardiovascular examination, even when done by a non-cardiologist, has a high negative predictive value and specificity, which suggests that the likelihood of significant valvular heart disease is low in the absence of a murmur.
- Clinical cardiovascular examination is recommended as the annual screening procedure for patients with prolactinoma taking cabergoline.
- Echocardiography is recommended for patients with an audible murmur; patients treated for more than 2 years at a dose of 3 mg or higher per week or equivalent (eg, up to 3 mg per week) and those remaining on cabergoline after age 50 years.

...group because our aim was not to compare the frequency of abnormalities between cases and controls, but to prospectively assess the usefulness of a screening cardiovascular examination in an endocrine clinic setting to rule out significant valvular pathology in patients with prolactinoma treated with cabergoline. To our knowledge, this is the first study to assess the utility of cardiovascular examination in this setting.

On the basis of our clinical data combined with data identified through our systematic review, we challenge the recommendation of routine annual echocardiography in all patients taking cabergoline for prolactinoma. In a low-risk asymptomatic patient with a normal cardiovascular examination, a clinician has a low probability of clinically significant valvular lesions by routine cardiovascular examination done during clinic visits. Patients on long-term treatment, taking high doses of cabergoline, or both, might eventually exceed a cumulative dose that could increase risk of cabergoline-associated valvulopathy. Therefore, we suggest an annual clinical cardiovascular examination for patients with prolactinoma taking cabergoline, and that an echocardiogram should be reserved for the following patient groups: those with an audible murmur on cardiovascular examination; those treated for 5 years at a dose of 3 mg or higher per week (or equivalent—eg, 1 mg per week for 15 years); and those who remain on cabergoline treatment after the age of 50 years (because of the increased prevalence of valvular lesions with age and a possible age-related effect in Parkinson’s disease; panel 2). The suggested 3 mg or higher per week for 5 years equates to a cumulative dose of 720 mg, which is a conservative approximation of the lower standard deviation of the mean cumulative dose associated with cabergoline-associated valvulopathy in patients with Parkinson’s disease (4035 mg (SD 3208)). In view of cabergoline being given once to twice per week in patients with prolactinoma (and rarely exceeding 3 mg per week), calculation of cumulative doses in clinical practice is time consuming so this parameter is more convenient for the clinician. Most premenopausal women with microprolactinomas will never need an echocardiogram with these proposed criteria. The optimum frequency of repeat studies in high-risk patients remains to be established, but some data suggest that the development of cabergoline-associated valvulopathy is rare during 2 to 5 years. Care should also be taken when interpreting echocardiogram results because results in the clinical setting will be unmasked and at risk of over-reporting. If valvular lesions are detected, then attempts should be made to distinguish cabergoline-associated valvulopathy from other causes and to associate these with symptoms and the results of clinical examination. Collectively, these findings have important ramifications for deciding whether to continue cabergoline treatment in patients with prolactinoma.

Declaration of interests
We declare no competing interests.

Acknowledgments
This research did not receive any specific grant from any funding agency in the public, commercial, or not-for-profit sector. This study was undertaken at St Vincent’s Hospital Melbourne. The corresponding author had full access to all the data in the study and had final responsibility for the decision to submit the paper.

References
2.3 Publication 2

The third case of cabergoline-associated valvulopathy: the value of routine cardiovascular examination for screening.

Caputo C, Prior D, Inder W.

The Third Case of Cabergoline-Associated Valvulopathy: The Value of Routine Cardiovascular Examination for Screening

Carmela Caputo, David Prior, and Warrick J. Inder

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A decade after the alarming association of cabergoline-associated valvulopathy (CAV) in Parkinson disease, only two confirmed cases have occurred in patients with prolactinoma. Routine screening for CAV by echocardiography has not proven to be of diagnostic utility, has several limitations, and is not widely practiced. We have previously highlighted the value of annual cardiovascular examination as a screening tool for CAV in patients with prolactinoma. We present a case, now the third confirmed case of CAV, to highlight the value of the cardiovascular examination. A 52-year-old woman with a 25-year history of macroprolactinoma had received multimodal treatment, including surgery, radiotherapy, and medical therapy. Her medical therapy initially consisted of bromocriptine, followed by cabergoline. The cabergoline dose was 6 mg weekly. In 2006, the cumulative dose was 3272 mg when an echocardiogram showed no evidence of valvular disease. A routine cardiovascular examination in the clinic detected a new murmur in 2016. The echocardiogram demonstrated new-onset mild to moderate aortic regurgitation, with a thickened and restricted valve consistent with CAV. The cumulative dose of cabergoline at that point was 4192 mg. Follow-up echocardiography at 6-month intervals showed progression to moderate to severe aortic regurgitation, which has since stabilized. Cabergoline therapy was weaned and stopped completely in April 2017. An annual cardiovascular examination is the best screening test for CAV and can change the course of a patient’s treatment. Echocardiograms should be reserved for patients with a new-onset cardiac murmur or a high cumulative dose of cabergoline.

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Freeform/Key Words: cabergoline, prolactinoma, valvular heart disease

Indisputable evidence exists for the causal relationship of ergot-derived dopamine agonists (bromocriptine and cabergoline) and cardiac valve disease in Parkinson disease related to high cumulative doses. During the past decade, several studies have examined the risk of valvulopathy in patients with prolactinoma taking cabergoline. In our 2015 systematic review, the risk was found to be extremely low (only two confirmed and one possible case), most likely owing to the lower cumulative doses used in treating prolactinoma [1]. At the time of that review, 19 studies and 2 case reports pertaining to cabergoline and valvular heart disease had been reported. Since then, three further reported studies have not shown an increased prevalence of cabergoline-associated valvulopathy (CAV): now totaling 2000 cases of prolactinoma studied [2–4]. Studies of serial echocardiograms in patients with prolactinoma have not shown new cases of CAV [2, 5, 6]; however, in reality, few patients are undergoing serial echocardiograms [5].

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Given the low prevalence of CAV in patients with prolactinoma, we believe that optimal screening should consist of an annual cardiovascular examination for all treated patients, with echocardiograms directed at those with a murmur or high cumulative doses of cabergoline.

We present the case of a patient to emphasize the value of the cardiovascular examination, because this is now the third confirmed case of CAV. The patient provided written informed consent for the reporting of her case.

1. Case Report

A 52-year-old woman had originally presented in 1989 at age 22 years with headaches, secondary amenorrhea, and galactorrhea. The prolactin level was elevated to >6000 mIU/L (undiluted), and a CT scan showed a 3-cm adenoma extending into the sphenoid sinus. She was initially treated with bromocriptine from 1990 to 1992 with some initial shrinkage to 2.5 cm on subsequent scans, with a prolactin level of 10,000 mIU/L (reference range <615 mIU/L) after serial dilution. Because of the limited response to bromocriptine with the lesion abutting the optic apparatus, she subsequently underwent two debulking microscopic transsphenoidal surgeries in 1992. However, her prolactin level remained elevated at 7000 mIU/L, with residual disease in the right cavernous sinus. Bromocriptine was started again, until cabergoline became available in 1997. Cabergoline was begun at 2 mg weekly and subsequently increased to 6 to 7 mg weekly over time, with failure to ever reduce the prolactin level to <1000 mIU/L. After initially declining radiotherapy in 1999, the patient subsequently underwent stereotactic radiosurgery in 2004 and continued with cabergoline at high doses.

The patient had panhypopituitarism and was taking cortisone acetate and thyroxine. She had long-standing hypogonadism but had been variably compliant with estrogen and progestin replacement. She developed osteopenia and experienced a low trauma fracture of the fibular in January 2017, thereafter beginning antiresorptive therapy.

The patient underwent echocardiography in 2009, which showed no evidence of valvular disease. The weekly dose of cabergoline was 6 mg, and the cumulative dose was 3272 mg. Her prolactin level at this time was at its nadir of 900 to 1000 mIU/L. In 2012, a repeat MRI scan had shown a substantial reduction in the residual adenoma, after which the dose of cabergoline had been weaned at the rate of 0.5 mg every 6 months.

A routine cardiovascular examination detected a new murmur in 2016. The patient was asymptomatic. The echocardiogram demonstrated new-onset mild to moderate aortic regurgitation, with a thickened and restricted valve—the three cardinal echocardiographic features of CAV. The cumulative dose of cabergoline at that point was 4192 mg, with a weekly dose of 3 mg. Follow-up echocardiograms at 6-month intervals showed progression to moderate to severe aortic regurgitation, which at the last follow-up examination had stabilized (Fig. 1). Cabergoline was progressively weaned and was stopped in April 2017. Her prolactin level increased to 4706 mIU/L, but no interval tumor growth was seen on MRI.

2. Discussion

Our patient's case represents the third confirmed case of CAV in >2000 cases of prolactinoma treated with ergot-derived dopamine agonists reported since 2008. Although other studies have shown evidence of various cardiac valvular abnormalities, CAV represents a specific pathology characterized by three salient echocardiographic features: (1) moderate or severe regurgitation, (2) valve thickening, and (3) valve restriction. Furthermore, the absence of calcification and myxomatous changes distinguishes CAV from age-related sclerosis and myxomatous valve disease, respectively. If a valvular lesion is found, it is crucial to distinguish among age-related changes, myxomatous changes, and CAV to determine whether cabergoline should be weaned or ceased. In our patient, the three salient features of CAV were present on the echocardiogram, leading to cessation of cabergoline therapy as
Figure 1. Echocardiographic images of a thickened and mildly restricted aortic valve (AoV) with resultant moderate to severe aortic regurgitation (AR). (a) Parasternal long axis views in which the AoV leaflets are mildly thickened and show mild doming owing to incomplete opening in systole. (b) The AoV leaflet tips are mildly thickened when the valve is closed. (c) Parasternal long axis view showing color Doppler image of a broad jet of AR almost filling the left ventricular outflow tract (LVOT). (d) Apical long axis view of the color Doppler jet of AR reaching the left ventricular apex. LA, left atrium; LV, left ventricle.

recommend by the Food and Drug Administration (FDA) [7]. Although measurement of the tricuspid valve tethering area and mitral valve tenting is useful in comparing defined patient groups to quantify the degree of valve restriction, these are subject to user and machine variability. They are not reliably reproducible and are only surrogate markers of subclinical changes in the valve [1]. In clinical practice, tethering and tenting are not echocardiographic markers that would change medical management.

On histologic examination, which is only available for those who have undergone surgical valve replacement, CAV is characterized by a thickened valve due to fibrous proliferation, with the absence of inflammatory cells, thrombus, and calcification [8]. The mechanism of valvular pathology as a cabergoline-induced adverse event is biologically plausible via stimulation of serotonin (5-hydroxytryptamine) receptor subtype 5-hydroxytryptamine(2B), which is expressed in heart valves and is known to mediate mitogenesis and proliferation of fibroblasts [9].

In a cohort of patients with Parkinson disease at risk of CAV, the mean cumulative dose was 4015 ± 3208 mg, giving one standard deviation less at ~720 mg [10]. Only 10% of patients with prolactinoma will require high doses of cabergoline (>3 mg/wk) [11]. Those
taking doses of 3 mg/wk will reach cumulative doses of 720 mg after 5 years of treatment. Most patients with prolactinoma will receive doses ≤1 mg/wk.

In the reported data, two cases of CAV in patients with prolactinoma have been confirmed. One case of CAV occurred with a cumulative dose of only 252 mg [8]. That patient had fulminant heart failure, with echocardiographic findings of severe mitral regurgitation and a thickened and restricted valve (in the absence of stenosis), confirming CAV. The histologic findings were available because the patient had required valve replacement, which supported the echocardiographic diagnosis. In the second case of CAV, moderate mitral regurgitation occurred at an extremely high dose of 5252 mg, similar to our case [12]. That patient also had all three features of CAV found on the echocardiogram; however, valve replacement was not required. One case of moderate tricuspid regurgitation has been reported; however, no morphological features of CAV were found on the echocardiogram, and the patient had only had a cumulative cabergoline dose of 48 mg. These findings together would make CAV less likely [13].

Our patient had been taking a high dose of cabergoline for many years, reaching doses comparable to those used for Parkinson disease. She had also received several years of bromocriptine, another ergot dopamine agonist. Despite normal echocardiographic findings when the cumulative dose was ~3000 mg, she subsequently developed CAV, which was initially detected clinically by the presence of a new murmur although she was asymptomatic.

A recent community study from the United Kingdom demonstrated that very few patients with hyperprolactinemia had undergone baseline echocardiography (2 of 45), and only 5 of the 45 patients had undergone serial echocardiograms 2 years apart [3]. Even for those with Parkinson disease, a study showed that very few patients had had serial echocardiograms [14]. Three studies have followed up a total of 185 patients with prolactinoma with serial echocardiograms after 2 to 6 years and did not find an increased incidence of CAV [2–4]. Thus, echocardiography is seldom used; however, more importantly, routine interval scanning has not been shown to be useful. In July 2011, the FDA recommended “regular echocardiograms every 6 to 12 months” in all users of cabergoline “or as clinically indicated with the presence of signs and symptoms such as dyspnea, edema, new cardiac murmur, or congestive heart failure” [7]. We would argue that the frequency of echocardiograms recommended by the FDA has no scientific basis. We have previously shown the limitations of echocardiography as a screening tool, in particular the risk of overreporting findings, failure to recognize the cardinal features of CAV (compared with age-related changes or myxomatous disease), and the associated financial and psychological costs [1].

In conclusion, it is reassuring to endocrinologists (and patients) that the prevalence of CAV in patients with prolactinoma is extremely low. Given this low prevalence, routine screening with echocardiography is not indicated. The recommended screening procedure should be an annual cardiovascular examination, with echocardiography reserved for patients with a murmur or those with high cumulative cabergoline doses.

Acknowledgments

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Disclosure Summary: The authors have nothing to disclose.

References and Notes


2.4 Publication 3

Letter to the editor: [A meta-analysis of the prevalence of cardiac valvulopathy in patients with hyperprolactinemia treated with cabergoline]

*Caputo C*, Inder W.


Letter to the Editor: [A Meta-Analysis of the Prevalence of Cardiac Valvulopathy in Patients with Hyperprolactinemia Treated with Cabergoline]

Carmela Caputo and Warrick J Inder

The Journal of Clinical Endocrinology & Metabolism
Endocrine Society

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Letter to the Editor: [A Meta-Analysis of the Prevalence of Cardiac Valvulopathy in Patients with Hyperprolactinemia Treated with Cabergoline]

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We are writing in response to the article published in The Journal of Clinical Endocrinology & Metabolism by Stiles et al., meta-analysis of the prevalence of cardiac valvulopathy in patients with hyperprolactinemia treated with cabergoline (1). The authors examined 13 case control studies, concluding that low dose cabergoline appears to be associated with increased prevalence of tricuspid regurgitation. Furthermore, “publication of large-scale, prospective, controlled, quality-assured echocardiographic studies with centralized standardized interpretation are needed to provide data and that additional cross-sectional studies drawn from routine practice are unlikely to inform the field significantly”. We disagree with this conclusion and suggest that meta-analysis is not the most appropriate means to determine the specific effects of cabergoline on cardiac valve morphology.

The conclusion that “cabergoline is associated with increased prevalence of tricuspid regurgitation warranting further studies” is misleading. The data presented for moderate or severe tricuspid regurgitation over 12 months of treatment were derived from 3 studies (Figure 4) and heavily influenced by one study, whose findings have never been replicated by others, nor even by the same group in their follow up study (2) (3). The purpose of a review should be to report on the drug specific condition of cabergoline associated valvulopathy (CAV) which is a specific condition defined by the echocardiographic triad of 1) moderate or worse regurgitation 2) valve thickening and 3) valve restriction in the absence of calcification, rather than report on the presence of regurgitation compared to an unexposed control group that is not associated with any other features of CAV (4). The need for a control group from the healthy population not taking cabergoline is unnecessary for detecting the disease specific condition of CAV, which can only occur in an exposed group. While understandable in terms of meta-analysis methodology, the specific exclusion of over 1000 cases published in cross sectional studies and long term follow up studies, is a major limitation.

To date there have only been 3 confirmed cases of CAV in prolactinoma patients. One occurred with a cumulative cabergoline dose of only 252mg (5). This case had severe mitral regurgitation, with a thickened and restricted valve (in the absence of stenosis), and with histological confirmation post valve replacement. The second case of moderate mitral regurgitation with all three features of CAV on echocardiography, occurred at an extremely high dose of 5252mg (6). The third case, with moderate to severe aortic regurgitation and all cardinal echocardiographic features of CAV, with cumulative dose of cabergoline of 4192mg.
(7). CAV occurring in the tricuspid valve in prolactinoma patients has not been reported to date.

CAV is a very uncommon complication of cabergoline treatment in the doses used for pituitary adenomas, which historically have been mainly prolactinomas. We reiterate our position that routine clinical examination by cardiac auscultation, with echocardiography reserved for patients with a new onset cardiac murmur or high cumulative drug exposure is the optimal method to screen for these rare cases and would encourage endocrinologists to report cases which fulfill the strict definition of CAV (4) (7).

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Disclosure Summary:
The authors have nothing to disclose.

References
2.5 Summary and conclusion

This chapter gives a chronological sequence of work I have done over the last few years in examining and understanding the risk of CAV in prolactinoma patients. It starts with examining the risk of CAV in a local Australian cohort, undertaking a systematic review of the literature and making recommendations about optimal screening (Publication 1), a case report (Publication 2) and a “Letter to the Editor” (Publication 3); these later two works supporting the original findings.

The results of this chapter have clarified several important clinical aspects that are of importance to the management of cases with prolactinoma who are prescribed cabergoline. Firstly, that the risk of CAV is in fact low, but is related to higher cumulative doses of cabergoline. Fortunately, very few (<10%) of cases require high doses of cabergoline of \( \geq 3 \text{mg per week} \). Cumulative doses above 720mg were associated with an increased risk of valvular heart disease in a PD cohort; a prolactinoma patient on 3mg per week would reach this dose after 5 years of treatment, and those on 1mg per week would reach this dose after 15 years. This information is certainly helpful as it changes management by consciously considering minimising lifetime exposure to cabergoline by using the lowest effective dose and for the shortest duration possible.

The findings from this chapter also clarify the diagnosis of CAV on echocardiogram, which had not been previously appreciated in the published studies which were reporting non-specific valvular abnormalities in prolactinoma patients, except for one study (49). I highlighted the specific findings on echocardiogram that need to be distinguished from other more common causes of valvular abnormalities. This has finally been highlighted in March 2019, in the first joint position statement by the British Society of Echocardiography, the British Heart Valve Society and the Society for Endocrinology for “monitoring patients receiving dopamine agonist therapy for hyperprolactinaemia” (45). The recommendations from this guideline however are for frequent echocardiographic screening with yearly scans.
for those on >2mg per week, and five yearly scans for those taking ≤2mg per week. They do not advocate the use of a cardiovascular examination on the basis there is a lack of evidence for its usefulness in being able to accurately diagnose an abnormality. The findings of my thesis argue against these recommendations. I discuss the limitations of echocardiograms and that there is a financial and psychological cost of such frequent scans. I show that there is evidence that a screening test of a cardiovascular examination is very effective at excluding a serious valve abnormality if no murmur is heard on auscultation.

These findings of this chapter have considerable impact on the management of all cases of prolactinoma who are starting or continuing cabergoline. This information needs to be discussed with all patients for informed consent who are taking this drug. The detection of a valvular abnormality on echocardiogram requires careful assessment to confirm the presence of CAV. The confirmation of CAV requires action with cessation of therapy and appropriate cardiological management. Without a doubt, the multidisciplinary pituitary team may need to be consulted in order to consider optimal alternative management of the prolactinoma. I continue to be interested in CAV in prolactinoma patients as even after a decade information is evolving and differences in opinion about how to optimally screen for CAV are still emerging.
Chapter 3: The risk of regrowth and recurrence in operated cases of non-functioning pituitary macroadenomas at a single Australian centre

3.1 Introduction

Surgery is the treatment of choice for NFPMAs which exhibit symptoms of mass effect including visual disturbance, headache and impaired pituitary hormone function. Unlike the case for prolactinomas, no effective medical therapies are available for NFPMAs. The natural history of most unoperated NFPMAs is to grow over time, gradually exerting mass effect on surrounding structures (58). A major issue with operating on NFPMAs is that surgery may not be curative with a long-term risk of regrowth and recurrence. Other major issues relate to hormone deficiencies and impairments to quality of life.

As discussed in the literature review in Chapter 1, the long-term outcome of NFPMAs following surgery has been considered unclear, with varying rates of regrowth and recurrence from data across the world. However, the data are consistent that the presence of residual disease is a strong risk factor for regrowth. At the time of publishing the findings of this chapter, there was only one small Australian study that reported on the prevalence of residual disease occurring post-operatively. Authors Bokhari et al., reported that in 39 cases of NFPMAs residual disease occurred in 51% (78). Clinical aspects that remain poorly defined are other risk factors for regrowth and recurrence, optimal management of residual disease and optimal duration of long-term radiological surveillance. No long-term surgical outcome data for NFPMAs had been available from Australia at the time of undertaking this study.
This chapter aims to describe the surgical outcomes of a large Australian cohort with NFPMAs examining the prevalence of residual disease post-operatively, the risk of regrowth and recurrence over time and comparing these to international series.

The study was undertaken by retrospective chart review of all surgical cases of NFPMAs at St Vincent’s Hospital Melbourne, Victoria, Australia from 1995-2013. This is a tertiary centre that undertakes a high volume of pituitary surgeries (approximately 40 cases per year) via expert pituitary neurosurgeons.
3.2 Publication 1

Younger age a risk factor for regrowth and recurrence of non-functioning pituitary macroadenomas: Results from a single Australian centre.


Younger age is a risk factor for regrowth and recurrence of nonfunctioning pituitary macroadenomas: Results from a single Australian centre

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Summary
Objective: The natural history of nonfunctioning pituitary macroadenomas (NFPMA) after surgical resection is variable, with guidelines unable to define the duration of radiological follow-up. In this first Australian series, we identify risk factors for regrowth/recurrence of NFPMA to assist with guiding recommendations for long-term follow-up.

Design: Retrospective analysis of all radiotherapy-naïve cases with NFPMA resected between 1995 and 2013.

Patients: One hundred and twenty-three cases had both ≥2 postoperative scans and ≥12-month follow-up.

Measurements: Regrowth was defined as any sustained increase in diameter of residual adenoma or recurrence as any new adenoma occurring post complete resection on serial pituitary MRI.

Results: Median follow-up time was 48 months (interquartile range [IQR]: 31-86). Overall regrowth/recurrence occurred in 29% (36/123). Regrowth occurred in 40% (30/76) at a median time of 44.5 months (IQR 22-80) compared to recurrence of 12.5% (6/48; P=0.003), occurring at a median time of 48 months (IQR 12-96; P=0.7). Further treatment was required in 66.7% and 56.7%, respectively (P=1.0). Risk factors for regrowth/recurrence by multivariate analysis were presence of residual disease and younger age at presentation. The longest time for regrowth was 168 months (14 years) and recurrence 156 months (13 years).

Conclusions: Presence of postoperative residual adenoma and younger age at presentation are the main predictors of regrowth/recurrence in NFPMA. Long-term serial imaging is required to detect regrowth and recurrence in younger patients and those with residual disease. Most regrowth/recurrences will occur within 10 years of follow-up.

KEYWORDS
pituitary macroadenoma, recurrence, regrowth
1 | INTRODUCTION

Trans-sphenoidal surgery is the treatment of choice for nonfunctioning pituitary macroadenomas (NFPA), which exhibit symptoms of mass effect including visual disturbance, headache and impaired pituitary hormone function. The natural history of most un-operated NFPA is to grow over time, gradually exerting mass effect on surrounding structures. The long-term outcome of NFPA following surgery has been considered unclear, with varying rates of regrowth and recurrence from data across the world. However, data are consistent that the presence of residual disease is a strong risk factor for regrowth. What remain poorly defined are other risk factors for regrowth/recurrence, optimal management of residual disease and optimal duration of long-term radiological surveillance.

Historically cases with residual disease were treated with postoperative radiotherapy to prevent regrowth. This practice developed due to postoperative regrowth rates of up to 75% seen when resection was performed via the trans-cranial approach. Some authors, including a recent guideline, continue to advocate this practice for those with residual disease. Postoperative radiotherapy is very effective at reducing regrowth rates, but there is an increased risk of de novo pituitary insufficiency, visual field deficits, cognitive dysfunction, cerebrovascular disease and possibly secondary intracranial tumours.

More recently, radiotherapy has been targeted to those considered to be at high risk of regrowth or recurrence, although what defines “high risk” is not well understood. Many series have documented the regrowth/recurrence rate for a subset of patients who did not receive postoperative radiotherapy as they were deemed low risk, introducing a significant selection bias. These studies of low risk populations report recurrence-free survival of 82%-90% at 5 years and 56% at 10 years. Studies documenting unselected radiotherapy-naive cases report 5-year recurrence-free survival rates of 99%-77%.

The optimal duration of radiological follow-up of operated NFPA remains poorly defined, with a recent systematic review and guideline being unable to conclude the duration of time or frequency of follow-up for those with or without residual disease, nor those given radiotherapy. We describe surgical outcomes and regrowth rate of NFPA from a single Australian centre and identify risk factors that may predict regrowth/recurrence, to assist with planning of long-term radiological follow-up. At our centre, secondary interventions such as postoperative radiotherapy or further surgery are reserved until regrowth or recurrence is demonstrated on radiological follow-up, this decision made by consensus opinion of the pituitary multidisciplinary team (MDT). This is the first published series from a single Australian centre.

2 | METHODS

All cases who underwent initial pituitary surgery for NFPA between April 1995 and December 2013 were reviewed. Ethics approval was obtained from the local Human Research Ethics Committee. Diagnosis of NFPA was based on the presence of a pituitary adenoma ≥1 cm in maximal diameter without biochemical evidence of hormonal hypersecretion confirmed by histological and immunohistochemical assessment.

Review of the medical histories was undertaken to obtain clinical, hormonal, ophthalmologic and radiological details. Visual fields were defined as normal or abnormal based on computerized visual field testing. Multiple hormone deficiency (MHD) was defined by the hypofunction of at least two individual hormone axes. Details and definitions of hormone deficiencies have been previously described.

Postoperative pituitary imaging was performed at 3 and 12 months and then annually or as clinically indicated. All cases were followed with serial MRI pituitary except four who had CT due to MRI contraindications. Pre- and postcontrast images were obtained. Scans were reported by different radiologists throughout the follow-up period but also reviewed by the primary neurosurgeon with the MDT of endocrinologists and radiation oncologists who reviewed the case notes and imaging for each patient as part of routine follow-up.

Initial adenoma was classified by Hardy classification and measured by the largest diameter in millimetres (mm). Operative resection was classified as complete or incomplete based on the presence of adenoma remnant on the first postoperative scan. Cases with regrowth or recurrence were treated on an individual basis with a combination of repeat surgery, radiotherapy or close observation, as recommended by the MDT.

Recurrence was defined as any new radiologically detected pituitary adenoma after initial complete resection. Regrowth was defined as a sustained increase in size of residual adenoma tissue in any dimension ≥2 mm detected by the reporting radiologist and confirmed by the MDT compared to the residual adenoma measured on the baseline postoperative scan.

2.1 | Statistical analysis

Results are expressed as mean±standard deviation (SD) for parametric variables and median, interquartile range (IQR) for nonparametric variables. Categorical variables were analysed using contingency tables and Fisher’s exact test. Continuous variables were analysed using the unpaired t test. Univariate and multivariate analyses were used for risk factors of regrowth/recurrence. Statistical significance was set at P<0.05, and data were analyzed using STATA 11.0; StataCorp, TX, USA.

Kaplan-Meier curves were generated to describe total time at risk. Lifetables were used to determine 5- and 10-years survival rates. Log rank tests and univariate Cox regression were used to compare the survival rates of each variable. Based on univariate analysis, residual size and location, immunohistochemical staining, apoplexy and surgery were excluded from multivariate analysis. Initial adenoma size was excluded from the multivariable model due to the number of missing observations. Age was analysed by quintiles with 41 and under being the youngest age group. This was included in the model as a binary variable (41 and under vs >41); however, similar results were produced if age was used as a continuous variable. Given the increased regrowth/recurrence likely to be seen with longer duration of follow-up, outcomes were adjusted for follow-up duration in the current study.
3 | RESULTS

A total of 177 (97 male) consecutive operated NFPMA cases were identified. The median age at diagnosis was 57 years (IQR: 45–69; Table 1). Three (1.7%) had immediate postoperative radiotherapy due to large postoperative remnant; these were excluded from the regrowth/recurrence analysis. In total, 123 cases (69.3%) had more than 12-month follow-up and at least two postoperative scans.

3.1 | Mode of presentation and hormonal status

Sixty-one percent (108/177) of cases presented due to symptoms of mass effect or hormonal dysfunction, 10.7% (n=19) presented with apoplexy, and the adenoma was discovered incidentally on imaging performed for other reasons in the remaining 28.2% (n=50, Table 1). 69% (118/177) of cases reported visual disturbance at baseline, of which 48.3% (57/118) had bitemporal hemianopsia. Postoperative visual improvement occurred in 88% (73/80).

Mean pre-operative prolactin concentration was 699±528 mU/L (range 14–3303 mU/L). Prolactin was >2000 mU/L in 2.7% (4/148) of cases. Over 58% of cases had at least one axis deficiency preoperatively, and more than one-third had MHD. At 6 months postoperatively, 38% had MHD (Table 1).

3.2 | Surgery

Surgery was performed by the primary surgeon (P McN) in 72% (127/177) of cases; the remainder being performed by one of five other neurosurgeons. The trans-sphenoidal (microscopic) technique was utilized in 161 (91%), trans-sphenoidal (endoscopic) in 15 (8%), and one case underwent craniootomy (1%). The majority 88.1% (156/177) underwent one procedure, 9.6% two procedures and 2.3% three procedures during the follow-up period (Table 1).

3.3 | Postoperative complications

One or more postoperative complications occurred in 52.2% (37/177) of cases at first surgery (Table 1). This included the syndrome of inappropriate antidiuretic hormone (SIADH) 8% (14), transient diabetes insipidus (DI) 15.3% (27), permanent DI 6.8% (12), cerebrospinal fluid (CSF) leak 4.5% (8) and cardiac events 1.1% (2). One patient needed removal of retained nasal packs (DI,1%). There were two cases of significant postoperative bleeding; one case on warfarin, who had a preoperative INR of 1.2, and the other case who required urgent surgery due to pituitary apoplexy postoperative artery bypass grafting (CABG) treated with tirolbin and intravenous heparin. There were three (1.7%) cases of CSF infection. The primary surgeon had a lower complication rate compared to the others combined (P=0.04), which was more marked if transient DI was excluded from the analysis (P=0.02).

There were 10 deaths during follow-up. One occurred within 3 months of operation in a 52-year-old female who developed pituitary apoplexy whilst thrombocytopenic in the setting of chemotherapy for acute myeloid leukaemia. She died 1 week postoperatively due to ventricular fibrillation during neutropenic sepsis. The remaining nine deaths occurred between 6 months and 9 years of follow-up: cause of death was unrelated malignancy (three cases), cerebrovascular accident (two cases) or unknown causes (four cases). Median age of death was 79 years (IQR: 68.5–82.5).

Immunohistochemistry was available for 174 adenomas. The majority, 46%, were null cell adenomas and 6.3% were silent ACTH adenomas (Table 1).

3.4 | Regrowth/recurrence

Analysis of adenoma regrowth or recurrence was undertaken on 123 cases. Of the original cohort (177), 54 cases were excluded; 34 had <12-month follow-up at our centre; 17 failed to attend their second postoperative scan; and three cases underwent immediate postoperative radiotherapy. Nine cases underwent repeat surgery due to stable large postoperative residual; these cases were not classified as having regrowth. Follow-up was until regrowth/recurrence was detected or most recent recorded MRI. Median follow-up time was 48 months (IQR 31–86). There was a similar duration of follow-up for those with and without regrowth/recurrence.

Residual adenoma was demonstrated in 61% (76/123) of cases on the first postoperative scan, with 40% (30/76) located intrasellar, 8% (6/76) suprasellar and 49% (37/76) involving one or both cavernous sinuses. The location of residual adenoma was unrecorded in two of 76 cases. Those where the initial scan was unclear as to the presence of residual adenoma were classified according to the result of the second postoperative scan.

The overall rate of regrowth and recurrence was 29% (36/123). Regrowth of adenoma was observed in 40% (30/76) at a median time of 44.5 months (IQR: 22–80) compared to recurrence of 12.5% (6/48; P=0.03), occurring at a median time of 48 months (IQR: 12–96).

There was no significant difference in the time to regrowth vs recurrence (P=0.7, Table 2). Two cases of adenoma growth were detected at >1 year follow-up; a regrowth at 168 months (14 years) and a recurrence at 156 months (13.3 years).

Overall 5- and 10-years progression-free survival rates were 77% and 50%, respectively. Progression-free survival amongst the cohort with no postoperative residual adenoma seen on initial scan was 92% at 5 years and 77% at 10 years compared to 67% and 38% in those with postoperative residual adenoma (P=0.07 and P=0.007, respectively, Table 2 and Figure 1).

In the 36 cases with regrowth or recurrence, intervention was required in 21 of 36 (58%), consisting of repeat surgery in 32% (n=7), radiotherapy in 38% (n=8) and both modalities in 29% (n=6). The remaining 42% (n=15) cases were observed, with further serial imaging planned. Rates of treatment and observation were similar between those with regrowth or recurrence (Table 2).

Significant factors associated with regrowth/recurrence after multivariate analysis were presence of postoperative residual adenoma (P=0.0008) and younger age at presentation (P=0.0003). Univariate analysis
<table>
<thead>
<tr>
<th>TABLE 1 Baseline demographical details</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Gender (%)</strong></td>
</tr>
<tr>
<td>Male</td>
</tr>
<tr>
<td>Female</td>
</tr>
<tr>
<td><strong>Age years (Median [IQR])</strong></td>
</tr>
<tr>
<td>Male</td>
</tr>
<tr>
<td>Female</td>
</tr>
<tr>
<td><strong>Presentation (%)</strong></td>
</tr>
<tr>
<td>Incidental</td>
</tr>
<tr>
<td>Symptomatic</td>
</tr>
<tr>
<td>Apoplexy</td>
</tr>
<tr>
<td><strong>Headache (%)</strong></td>
</tr>
<tr>
<td>69/177 (40), 69/108 (63.8) symptomatic cases</td>
</tr>
<tr>
<td><strong>Loss of libido/impotence males (%)</strong></td>
</tr>
<tr>
<td>29/97 (29.9)</td>
</tr>
<tr>
<td><strong>Oligo/Amenorrhoea (%)</strong></td>
</tr>
<tr>
<td>25/30 (83.3)</td>
</tr>
<tr>
<td><strong>Galaactorrhoea females (%)</strong></td>
</tr>
<tr>
<td>10/32 (31.3)</td>
</tr>
<tr>
<td><strong>Visual abnormality (%)</strong></td>
</tr>
<tr>
<td>138/171 (69)</td>
</tr>
<tr>
<td><strong>Surgical technique (%)</strong></td>
</tr>
<tr>
<td>Microscopic</td>
</tr>
<tr>
<td>Endoscopic</td>
</tr>
<tr>
<td>Transcranial</td>
</tr>
<tr>
<td><strong>Number of surgeries (%)</strong></td>
</tr>
<tr>
<td>1 surgery</td>
</tr>
<tr>
<td>2 surgeries</td>
</tr>
<tr>
<td>3 surgeries</td>
</tr>
<tr>
<td><strong>Hormonal deficiency status</strong></td>
</tr>
<tr>
<td>Pre-operatively</td>
</tr>
<tr>
<td>65/108 (60.2)</td>
</tr>
<tr>
<td><strong>Thyroid</strong></td>
</tr>
<tr>
<td>39/121 (32.2)</td>
</tr>
<tr>
<td>38/115 (33)</td>
</tr>
<tr>
<td><strong>Adrenal</strong></td>
</tr>
<tr>
<td>30/119 (29.4)</td>
</tr>
<tr>
<td>46/116 (39.6)</td>
</tr>
<tr>
<td><strong>Permanent DI</strong></td>
</tr>
<tr>
<td>0/177 (0)</td>
</tr>
<tr>
<td>12/177 (6.8)</td>
</tr>
<tr>
<td><strong>Multiple (62)</strong></td>
</tr>
<tr>
<td>42/120 (35)</td>
</tr>
<tr>
<td>43/112 (38)</td>
</tr>
<tr>
<td><strong>Surgical complications</strong></td>
</tr>
<tr>
<td>Total 177 (%)</td>
</tr>
<tr>
<td>NdN 127 (%)</td>
</tr>
<tr>
<td>Others 50 (%)</td>
</tr>
<tr>
<td><strong>Total cases with ≥1 complications</strong></td>
</tr>
<tr>
<td>57 (32.2)</td>
</tr>
<tr>
<td>38 (29.9)</td>
</tr>
<tr>
<td>23 (46)</td>
</tr>
<tr>
<td><strong>Immunohistochemistry (%)</strong></td>
</tr>
<tr>
<td>NCl</td>
</tr>
<tr>
<td>Mixed</td>
</tr>
<tr>
<td>Pure LH/FSH</td>
</tr>
<tr>
<td>Silent ACTH</td>
</tr>
<tr>
<td>Necrotic</td>
</tr>
<tr>
<td>TSH</td>
</tr>
<tr>
<td>PRL</td>
</tr>
<tr>
<td>GH</td>
</tr>
</tbody>
</table>

**CSF, cerebrospinal fluid; DI, diabetes insipidus; IQR, Interquartile range; %, percentage; SIADH, syndrome of inappropriate antidiuretic hormone.**
TABLE 2  Characteristics of those with regrowth/recurrence

<table>
<thead>
<tr>
<th></th>
<th>Regrowth if residual adenoma (N=30)</th>
<th>Recurrence if no residual adenoma (N=6)</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Time to growth (mo)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Median (IQR)</td>
<td>46 (24-84)</td>
<td>48 (12-96)</td>
<td>.7</td>
</tr>
<tr>
<td>Progression-free survival, %</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>5 yr</td>
<td>67</td>
<td>92</td>
<td>.07</td>
</tr>
<tr>
<td>10 yr</td>
<td>38</td>
<td>77</td>
<td>.007</td>
</tr>
<tr>
<td>Gender (%)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>21/40 (52.5)</td>
<td>4/31 (12.9)</td>
<td>.001</td>
</tr>
<tr>
<td>Female</td>
<td>9/35 (25.7)</td>
<td>2/17 (11.8)</td>
<td>.47</td>
</tr>
<tr>
<td>Recurrence/regrowth</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>requiring intervention</td>
<td>17/75 (22.6)</td>
<td>4/48 (8.3)</td>
<td>.67</td>
</tr>
<tr>
<td>Intervention (%)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Observation</td>
<td>13/30 (43.3)</td>
<td>2/6 (33.3)</td>
<td>1.00</td>
</tr>
<tr>
<td>Intervention</td>
<td>17/75 (22.6)</td>
<td>4/48 (8.3)</td>
<td>.67</td>
</tr>
<tr>
<td>Surgery alone</td>
<td>7/23 (30.4)</td>
<td>0/6 (0)</td>
<td>.01</td>
</tr>
<tr>
<td>Radiotherapy alone</td>
<td>5/16 (31.2)</td>
<td>2/5 (40)</td>
<td>.18</td>
</tr>
<tr>
<td>Surgery &amp; radiotherapy</td>
<td>5/16 (31.2)</td>
<td>1/6 (16.7)</td>
<td>.21</td>
</tr>
</tbody>
</table>

IQR, Interquartile range.

*P-value for observation v intervention.

FIGURE 1  Kaplan-Meier survival for regrowth/recurrence in those with and without residual disease. Progression-free survival. Analysis time in months. CI, confidence interval

Indicated that there was a regrowth rate reduction of approximately 3% for every year older a subject was at presentation. The rate of regrowth remained 4.2 times higher in individuals 41 and younger compared to those aged over 41, after accounting for gender and the presence of residual disease (Figure 2). The rate of regrowth remained 4.8 times greater than recurrence, after accounting for gender and age (Figure 3).

Larger baseline adenoma size was a predictor of regrowth/recurrence (P=0.05, Table 3); however, it was not included in the multivariate analysis as there were 19 cases where these data were missing. However, this association may still be significant given the likelihood that the cause for the missing data was random.

FIGURE 2  Kaplan-Meier survival for regrowth/recurrence by age. Analysis time in months. CI, confidence interval

On univariate analysis, males with residual disease were significantly more likely to regrow than females with residual disease (32.5% vs 23.7%, P=0.03), but the effect of gender was not significant when examining regrowth and recurrence combined (P=0.10; Table 2). There was no association with risk of regrowth/recurrence and presence of extracellular adenoma at baseline (P=0.4), extracellular residual (P=1.0; Table 3), size of residual adenoma (P=0.2) nor presentation with apoplexy (P=0.3; Table 3). Regrowth/recurrence requiring treatment was strongly predicted by extracellular adenoma at baseline (P=0.01), but not extracellular residual (P=0.4) nor apoplexy (P=1.0).

Adenomas with invasion into one or more cavernous sinuses were less likely to be completely resected (P=0.01); however, this did not
TABLE 3  Determinants of regrowth/recurrence

<table>
<thead>
<tr>
<th></th>
<th>Regrowth/recurrence (N=36)</th>
<th>No regrowth (N=87)</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Residual</td>
<td>30</td>
<td>45</td>
<td>.0008</td>
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<tr>
<td>No residual</td>
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<tr>
<td>Median (IQR)</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Overall</td>
<td>50 (39-62)</td>
<td>57 (46-67)</td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>54 (34-63)</td>
<td>57 (46-66)</td>
<td></td>
</tr>
<tr>
<td>Female</td>
<td>47 (39-63)</td>
<td>57 (46-67)</td>
<td></td>
</tr>
<tr>
<td>Binary ≤ 61; &gt;61</td>
<td></td>
<td></td>
<td>.0001</td>
</tr>
<tr>
<td>Adenoma size</td>
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<tr>
<td>Pre-operatively</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Median (IQR)</td>
<td>30 (23-36)</td>
<td>26 (20.5-33)</td>
<td>.05*</td>
</tr>
<tr>
<td>Duration of follow-up</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Median (IQR)</td>
<td>44.5 (21.5, 78)</td>
<td>52 (32, 100)</td>
<td></td>
</tr>
<tr>
<td>Gender</td>
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<td>Female</td>
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<td>41</td>
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<tr>
<td>Location of residual adenoma</td>
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<td>Intracapsular</td>
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<td>Immunohistochemistry</td>
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<td>Mixed</td>
<td>9</td>
<td>19</td>
<td></td>
</tr>
<tr>
<td>Pure LH/TSH</td>
<td>7</td>
<td>12</td>
<td></td>
</tr>
<tr>
<td>Silent ACTH</td>
<td>3</td>
<td>5</td>
<td>.76</td>
</tr>
<tr>
<td>Other (Necrotic, TSH, PRL, GH)</td>
<td>0</td>
<td>13</td>
<td></td>
</tr>
</tbody>
</table>

IQR, interquartile range; Mm, millimetres.

*p value calculated using Fisher's exact test, not log rank test.

predict regrowth or lack thereof. There was no difference between surgeons in terms of risk of residual adenoma (P= .5) nor postoperative regrowth/recurrence (P=.10).

There were 13 cases with apoplexy. None of the eight apoplexy cases without residual disease had recurrence; however, regrowth was observed in two of five with apoplexy who had residual adenoma.

Immunostaining for ACTH was not associated with increased risk of regrowth/recurrence (P=.76), although numbers were small.

4 | DISCUSSION

In this first Australian series of operated NPPMA, we have described the natural history of regrowth/recurrence in an unselected cohort of radiotherapy-naive cases. The two major risk factors are the presence of residual disease and younger age at diagnosis. Other studies have demonstrated that residual disease strongly predicts adenoma regrowth. An association between younger patient age and risk of regrowth/recurrence has been shown by some investigators, although not all.

Even in expert hands, the presence of residual disease is a frequent finding post-surgery. Single-centre series of NPPMA throughout the world report rates of residual disease between 54%–80% cases, similar to our finding of 61%, 3,5,11,13,15,16,18. An exceptionally low rate of residual disease of 32% was achieved by a recent Korean group using the traditional microscopic technique.

There is inconsistency in the rate of regrowth and recurrence in radiotherapy-naive series throughout the world literature. Series from Europe, Korea, and Israel report regrowth rates between 42% and 59% over mean of 4–6 years of follow-up and recurrences in 6.9%–20%. Notably, the lowest rates of regrowth/recurrence observed over long follow-up periods occurred in an Irish, Dutch, and North American series, with regrowth of 33.5%, 14% and 28%, and recurrences of 0% (0/26), 0% (0/27) and 5.2% (13/248), respectively. A French series by Sotos-Ones et al. 13 also reported low rates, but this could be explained by a shorter mean follow-up period of 2.3 years (32.2% regrowth and 0% (0/7) recurrences).

Invasive nature of adenoma at baseline is a predictor of regrowth/recurrence in some studies. This may be the case as those with larger, more invasive and possibly more aggressive adenomas at baseline are likely to have a more extensive remnant. Unlike the study by Greenman et al., pre-operative adenoma invasion into the cavernous sinuses did not predict regrowth/recurrence in the present study; however, it was associated with risk of postoperative residual adenoma. Given postoperative remnant is a well-described risk factor for regrowth, the association in those with invasive adenomas at baseline may, therefore, more accurately reflect the likelihood of residual disease in these subjects.

Some authors argue that the volume of remnant predicts adenoma regrowth. Extent and location of remnant may be a marker of an aggressively growing adenoma, which is more likely to regrow. However, in our series neither size (measured as maximal adenoma diameter) nor location of residual adenoma was predictors of regrowth/recurrence. Our findings would suggest that having any residual, irrespective of location, has the propensity for regrowth.

In our series, the majority of surgeries were undertaken by one surgeon. We did not find any difference in the prevalence of residual disease, nor the risk of regrowth/recurrence between the primary surgeon compared to the other surgeons.

In our series, younger age of diagnosis was a significant factor predicting regrowth/recurrence. We demonstrated a 3% increase in relative risk of regrowth/recurrence for every year a younger patient was at the time of presentation, meaning there was a lower risk of regrowth/recurrence in patients presenting with NPPMA later in life.

The older a patient was at time of presentation, the less likely they were to suffer regrowth/recurrence. This observation allows clinicians greater confidence when electing to decrease or cease surveillance in the elderly. Four European studies have found younger age to be a
risk factors, but this was not substantiated in the Korean, North American, nor two European series.

On univariate analysis, we found that amongst those with residual adenoma, males were more likely to exhibit regrowth than females. This association has not been found by others, and contrary to our findings, Park and colleagues found an increased risk of regrowth in female cases on univariate analysis.

A previous study showed immunohistochemical staining for ACTH was associated with more aggressive recurrence but not increased recurrence rates. In contrast, a more recent and larger study has shown a subgroup of silent corticotroph adenomas do have a higher recurrence rate. We did not demonstrate an association between ACTH staining and propensity for regrowth/recurrence; however, numbers were small. Mixed staining was not associated with any increase in regrowth/recurrence, in contrast to another series which reported an increased propensity for regrowth/recurrence in those with mixed staining and those who did not stain positive for gonadotrophins.

In this study, we have not reported proliferative marker Ki67 expression or p53 immunoreactivity, as these have only been utilized in recent years at our centre. The 2004 World Health Organisation classification of pituitary adenomas includes invasive growth, increased mitotic activity, high p53 immunoreactivity and Ki67 labelling index above 3%. Pituitary adenomas can constitute up to 15% of pituitary adenomas; however, the utility of these markers is still unclear in typical pituitary adenomas.

Pituitary apoplexy may have a lower risk of regrowth/recurrence possibly relating to apoplexy predicting the absence of residual adenoma. Although not reaching statistical significance due to small numbers, our data support that the presence of any residual disease following apoplexy remains a risk factor for regrowth. This is also supported by data from Oxford, where regrowth occurred in 3 of 27 cases of apoplexy, with all three having residual disease.

A pertinent clinical question posed by Reddy et al. is when to stop imaging those with operated NFPVA who are not irradiated. The authors were unable to define a time point from their study but adenoma growth occurred in cases after 10 years. They therefore concluded that imaging should continue indefinitely and recommended that early irradiation should be considered in those with postoperative extraosellar adenomas. The French Endocrine Society’s recent consensus paper addresses the frequency of follow-up imaging, suggesting annual imaging for the first 5 years then every 2-3 years in a case-dependent manner in those with residual disease. In those without residual disease, imaging should occur annually for the first 5 years and then at 7, 10 and 15 years, before ceasing. In our series, the longest recurrence after complete resection occurred at 13 years, but Reddy et al. reported 20% of recurrences occurring after 10-years follow-up, including one case at 25 years. Regardless of the presence of residual disease, we would advocate the indefinite scanning of those diagnosed at or before age 40. It must be highlighted that in all published series, the majority of cases with regrowth/recurrence will occur within 10 years of surgery.

Although extremely effective at preventing regrowth, we do not advocate routine postoperative radiotherapy for cases with residual adenoma, as half of cases will have been unnecessarily exposed to longer term risks. There is emerging interest in the use of the dopamine agonist, cabergoline, in preventing regrowth, with small, nonrandomized studies (totaling approximately 100 cases) reporting regrowth rates of approximately 14% in those treated with cabergoline compared to 42% of those not treated. In these studies, doses of cabergoline varied from 5 to 3 mg weekly, with variable follow-up between 6 months and 6 years. Given approximately half of cases with residual disease will regrow, half of whom require a second intervention, interest in cabergoline for prevention of regrowth will continue. Prospective randomized studies are required to address this clinical question.

The main limitations of our study are inherent to its retrospective nature. We were able to follow-up 70% of the operated cohort. Some cases were inevitably lost to follow-up or followed up by private practitioners; however, this is common to most series. We analysed adenoma size by maximum adenoma diameter instead of volume. Recently, it has been shown that volumetric measurement correlates with maximal adenoma diameter. Currently, this technique is not widely used clinically, but shows promise to calculate growth rates which may predict need for intervention.

Regrowth/recurrence of NFPVA after surgery is more likely in the presence of residual disease and in those who are younger at time of presentation. Males with residual disease are at increased risk of regrowth. We advocate lifelong imaging for those diagnosed at ≥1 years of age, irrespective of the presence of residual disease. In most cases that have shown regrowth or recurrence, initial observation is possible, but MRI discussion should guide further management.

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Nothing to declare.

Disclosure
The authors report no conflict of interest concerning the materials or methods used in this study or the findings specified in this article.

References

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3.3 Summary and conclusion

This study is the largest reported Australian cohort of surgically treated NFPMAs undertaken at a single tertiary centre between 1995-2013. It was found that residual disease after operative therapy is common occurring in 61% of 123 cases. These local results are in keeping with international series and demonstrates that local surgical outcomes are on par with the rest of the world. In two small Australian studies also from single centres, authors Bokhari et al., had reported residual disease in 51% of 39 cases of NFPMAs (78) and in a publication appearing after my publication, authors Davis et al., reported residual disease in 60.5% in 38 cases of NFPMA, (79). In my study, 40% of cases with residual disease developed regrowth over a median of 4 years, but in those with complete removal of adenoma, recurrence occurred in only 12.5%. In those with regrowth and recurrences, half required further intervention in the form of repeat surgery, radiotherapy or both modalities. From a clinical standpoint these data demonstrate a major problem faced by patients with NFPMAs, the long-term threat of uncured disease.

Larger baseline adenoma size was a predictor of regrowth and recurrence (p=0.05), however this parameter was not included in the multivariate analysis as there were 19 cases where data of adenoma size were missing. However, this association is highly likely to be significant given that the reason for the missing data was a random occurrence. In a multivariate analysis, it was found that risk factors for adenoma regrowth and recurrence included the presence of residual disease and younger age at presentation (especially if aged under 41 years). Based on this finding, I have recommended that young patients should be considered for lifelong radiological follow-up. In the literature, there is no consensus on the duration of radiological follow-up of cases of NFPMAs.

Research is now focusing on understanding mechanisms and pathways in pituitary adenoma tumorigenesis and identifying biomarkers that may be able to distinguish different pathophysiological behaviours of pituitary adenomas. Identifying pathways or biomarkers
that could reliably identify adenomas at increased risk of regrowth or recurrence would be of clinical benefit in assisting with management and determining frequency and duration of long-term follow-up or indeed be targets for treatment with medical therapies. The biomarkers of proliferation Ki-67 and p53 immunoreactivity are widely described in pituitary adenomas but these markers have actually not been shown to reliably predict adenoma invasiveness nor regrowth or recurrence (196) (197). In one study, elevated Ki-67 was associated with younger age but in several other studies an inverse relationship was found between elevated Ki-67 and age (198). Immunostaining for Ki-67 and p53 are widely available, however, this was not assessed in my study as immunostaining for these proliferative markers are not routinely undertaken at the study centre.

Several other biomarkers are currently under investigation. Pituitary tumor transforming gene (PTTG) is an oncogene that has been shown to be overexpressed in pituitary adenomas. There does appear to be an association with increased PPTG expression in invasive adenoma compared to non-invasive adenomas, but data is not consistent with regards to PTTG expression and risk of regrowth and recurrence, nor regarding age or gender (198) (199). Other areas being explored are abnormalities in signalling pathways of cell growth such as transforming growth factor beta (TGFβ), aberrant expression of epidermal growth factor (EGF) and its receptor (EGFR) and increased expression of angiogenic factor vascular endothelial growth factor (VEGF) and its receptor (VEGFR) (200). Further interest has emerged in small non-coding RNA, miRNA, which can negatively regulate post-transcriptional gene expression. Studies have shown that certain miRNA can be either over-expressed or down regulated in pituitary adenomas compared to normal pituitaries; studies have linked abnormal expression of miRNA to pathways of onco-suppressive activity, cell cycle restraints, regulating TGFβ signalling pathway and mitotic cell inhibitor of Wee1 (200). Currently in clinical practice however, there are no biomarkers that that have been shown to be of use in understanding the variable behaviour of NFPMAs.

The findings from this chapter have major impacts on the management of NFPMAs. From a local perspective these data are relevant for communicating and consenting of patients with
NFPMA for operation as it clarifies the expected outcome from surgery at the local centre. Secondly, these data highlight that operating on smaller adenomas is advantageous and is a consideration for the multidisciplinary team when discussing optimal timing for surgical intervention. From an international perspective, where recommendations for long-term radiological follow-up of NFPMAs post-surgery has not been defined, the findings of this study suggest that young patients, irrespective of the presence of residual adenoma, warrant indefinite radiology surveillance due to a higher risk of regrowth and recurrence.
Chapter 4: Hormonal outcomes in operated cases of non-functioning pituitary macroadenomas at a single Australian centre

4.1 Introduction

The second of the major issues pertaining to NFPMA s undergoing surgery relates to hormone deficiencies resulting from damage to the normal pituitary gland either before or after surgery. The clinical importance of pituitary hormone deficiencies is that patients require lifelong endocrinological follow-up, it may reduce quality of life and have major implications for issues such as fertility and pregnancy.

As discussed in the literature review in Chapter 1, NFPMA s have the highest rate of hormonal disturbances but no data exist examining potential gender differences. It has been observed that gonadal deficiency is common in both males and females but deficiencies of other pituitary hormones (thyroid and adrenal axes) are usually reported with genders combined. It is thus unclear whether there are specific gender differences in hormonal status at presentation and post-operatively in patients with NFPMA s. Identifying gender differences in hormonal outcomes are of clinical importance as hormonal replacement regimens differ between gender, especially related to menopausal status and fertility in females.

In this chapter, the hormonal outcomes of this first Australian cohort of NFMAs have been described. It explores hormone function, both at presentation and post-operatively, in those with NFPMA s and seeks to determine differences between gender; differences in gender have not been previously explored. Further understanding hormonal deficiencies in NFPMA s
may have major impacts on understanding and improving the outcomes for patients with NFPMAs.

The study was undertaken by retrospective chart review of all surgical cases of NFPMAs at St Vincent’s Hospital Melbourne, Victoria, Australia from 1995-2010. This is a tertiary centre that undertakes a high volume of pituitary surgeries (approximately 40 cases per year) via expert pituitary neurosurgeons.
4.2 Publication 1

Gender differences in presentation and outcome of non-functioning pituitary macroadenomas.

C Caputo, T Sutherland, S Farish, P McNeill, KW Ng, WJ. Inder.

Clinical Endocrinology. 2013, 78(4):564-70
Gender differences in presentation and outcome of nonfunctioning pituitary macroadenomas

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Summary

Objectives Few data exist regarding gender differences in hormonal outcomes in nonfunctioning pituitary macroadenomas (NFPMA). The aim was to assess whether there are gender differences in hormonal outcomes in NFPMA following pituitary surgery at a single centre.

Design and methods Retrospective review of cases undergoing a first surgical procedure for NFPMA. Preoperative hormonal function was available for 122 cases at presentation and 94 cases 6 months postoperatively. Multiple hormone deficiency was defined as ≥2 hormonal axes loses. Tumour size and invasion on MRI scan were assessed independently by a single neuroradiologist.

Results At presentation, men were more likely than women to have multiple hormonal deficiency (47% vs 28%, P = 0.038). Premenopausal women tended to have smaller adenomas than men, but neither adenoma size nor invasion was associated with multiple hormonal deficiency at presentation. Postoperatively, differences were observed with only 14% of premenopausal women exhibiting multiple hormone deficiency, compared with 36% of postmenopausal women and 46% of men (P = 0.03). Overall, postoperative hormonal recovery was observed in over one-third of cases. Greatest recovery occurred in the gonadal axis of 60% (6/10) premenopausal women compared with 19% (8/43) of other groups combined (P = 0.007).

Conclusions Premenopausal women with NFPMA appear to have favourable hormonal outcomes. This may be due to a complex interplay between smaller tumour size and shorter disease duration. There should be no hesitation in offering pituitary surgery to premenopausal women with NFPMA, who have the most to gain in terms of restoration of hormonal function.

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Introduction

Pituitary adenomas are the commonest intracranial neoplasm, with prevalence studies showing approximately one in 1000 individuals having a clinically relevant pituitary adenoma.1 In contrast, 10% of the population may harbour small pituitary adenomas detectable on routine neuroimaging.2 Recently, it has been shown that incidence rates increase with age are higher in women early in life but more common in men later in life.3 Furthermore, men tend to have larger tumours, possibly due to delayed diagnosis.4,5

Pituitary adenomas may be associated with hormone deficiency of varying degrees, with functioning tumours and macroadenomas having fewer deficiencies than nonfunctioning pituitary macroadenomas (NFPMA).6–10 NFPMA have the highest rate of hormonal disturbances, but few data exist examining gender differences. It has been observed that gonadal deficiency is common in both men and women.14–17 Deficiencies of other pituitary hormones (thyroid and adrenal axes) are usually reported with genders combined and include cases of pituitary apoplexy. It is thus unclear whether there are specific gender differences in hormonal status at presentation and postoperatively in patients with NFPMA.

The aim of this study was to determine whether such gender differences exist in hormonal function, both at presentation and postoperatively among subjects with NFPMA via a retrospective chart review. In addition, relationships between age at presentation, adenoma size and invasiveness, and hormonal status both pre- and postoperatively have been examined.

Patients and methods

Between August 1995 and October 2010, 141 consecutive cases of NFPMA were identified who underwent their first pituitary surgery at St Vincent’s Hospital, Melbourne, Australia. One case had a craniotomy, but the remainder were operated via the transphenoidal route. Surgery was undertaken by principal surgeon PM in 77% cases, and the remainder by four other surgeons.

Sixteen cases presented with pituitary apoplexy and were excluded from hormonal analysis. Preoperative hormonal data were available at presentation for 122 cases, at 6 months for 94 cases, and for paired pre- and postoperative hormones for 93 cases.
Definitions of hormone deficiency

Multiple hormone deficiency was defined by the hypofunction of at least two individual hormone axes. Gender subgroups considered were men, premenopausal women and postmenopausal women.

Secondary hypogonadism was defined in men as an early morning testosterone <8 nmol/l, in premenopausal women with a clinical history of oligomenorrhea or amenorrhea associated with low estradiol and in postmenopausal women with appropriately low gonadotrophins for age (<20 mIU/ml). Gonadotrophin deficiency postoperatively at 6 months included the addition of commencement of testosterone replacement in men and failure of return of menses in premenopausal women.

Secondary hypothyroidism was defined as a free T4 below the reference range associated with an inappropriate or low TSH, having excluded those previously on thyroid hormone replacement for primary causes.

Secondary adrenal insufficiency at presentation was diagnosed by the presence of any one of the following: commencement of steroid replacement preoperatively, early morning cortisol <250 nmol/l (highest recorded cortisol 159 nmol/l), or failure of cortisol to rise to >550 nmol/l during a 250 μg ACTH 1-24 (Synacthen, Novartis, Basel, Switzerland) test or insulin tolerance test. Postoperatively, definition of adrenal axis deficiency also included those requiring cortisol replacement at 6 months, as assessed by the treating endocrinologist based on repeated early morning cortisol or dynamic testing.

Prolactin was considered raised if above the reported gender-specific reference range of the individual assay used. Replacement of human growth hormone in adults is not funded in Australia. As a result, few cases had routine dynamic testing of the GH-IGF-1 axis; therefore, data on the GH-IGF-1 axis have not been reported.

Throughout the study period, several biochemical hormone assays were used with some cases having biochemical tests at private pathology laboratories; therefore, normal values were taken as per the laboratory reference range for the particular assay.

MRI scans were reviewed by a single neuroradiologist (TS), with measurements of maximal diameters in millimetres, and invasion defined as cavernous sinus involvement or breach of bone margins. All adenosmas were confirmed as NFPMAs by histology and immunohistochemical staining.

Ethical approval was obtained through the institutional Human Research Ethics Committee.

Statistics

Results are expressed as mean ± standard error of the mean (SEM). Chi-squared test was used for the comparison of two groups (women compared with men, premenopausal women compared with others combined, premenopausal women compared with men). To determine relationship between multiple hormone deficiency and continuous variables such as age and adrenoma size, mean comparison tests were used. Multiple hormone deficiency was adjusted for gender (women and men), age and tumour size using multiple logistic regression. Statistical significance was considered at P < 0.05, utilising statistical package Stata 11.0; StataCorp, TX, USA.

Results

Presenting features

In total, 141 cases of NFPMAs (including the 16 with pituitary apoplexy) underwent their first pituitary surgery: 76 men (mean age 57.8 years, range 19 85), 41 postmenopausal women (mean age 64.5 years, range 48 85) and 24 premenopausal women (mean age 42.3 years, range 27 52). Clinical presentation included visual disturbance in 68%, headache in 34%, apoplexy in 11% and incidentally discovered in 31%. Incidentalomas occurred significantly more frequently in postmenopausal women compared with premenopausal women and men (46% vs 17% and 26%, respectively, P = 0.009). No gender subgroup difference was found between reporting of headache, visual disturbance or apoplexy: Menstrual disturbance occurred in 18/23 (78%) premenopausal women, and sexual dysfunction was recorded in 19/76 (25%) men. Patients with apoplexy were excluded from subsequent analysis.

Tumour size tended to be larger in men compared with premenopausal women (30.3 ± 1.2 cm vs 25.7 ± 2.0 cm, P = 0.056) but not different compared with postmenopausal women (29.1 ± 2.0 cm, P = 0.6). Tumour size was significantly larger in men of equivalent age to premenopausal women (33.9 ± 2.6 cm vs 25.7 ± 2.0 cm, P = 0.02).

The presence of invasion did not differ between groups (46% men, 62% postmenopausal women and 46% premenopausal women, P = 0.27). Age was not associated with tumour size (P = 0.59) nor the presence of tumour invasion (0.22) at presentation.

Cases with incidentalomas were older than those presenting symptomatically: 63.4 ± 2.2 vs 54.5 ± 1.5 years, P < 0.005. This was skewed by the higher incidence in postmenopausal women. Incidentally discovered tumours were significantly smaller than those presenting symptomatically (25.7 ± 1.4 cm vs 30.8 ± 1.2 cm, respectively, P = 0.01).

Preoperative hormonal data

Hormonal data were reviewed in 122 cases: 61 men, 38 postmenopausal women and 23 premenopausal women (Fig. 1a). The relative frequency of individual hormone deficiencies in the cohort was gonadal (67%) > thyroid (39%) > adrenal axis (24%) with no significant difference in frequency between the gender groups apart from thyroid hormone deficiency which was more frequent in men. Men were more likely than women to exhibit multiple hormone deficiency at presentation (47% vs 28%, P = 0.038).
Univariate analysis for the presence of multiple hormone deficiency at presentation was not related to tumour size, age at presentation nor the presence of adenoma invasion (Table 1). When preoperative multiple hormone deficiency was adjusted for gender, age and tumour size, gender was no longer significant ($P = 0.25$).

Proactin was elevated above the gender-specific reference ranges in 68% of premenopausal women, 68% of postmenopausal women and 46% of men. Proactin levels were below 2000 mlU/l in all but four cases, with highest proactin reported in a man at 3303 mlU/l, falling to a nadir of 1481 mlU/l postoperatively but subsequently rising to 2591 mlU/l during follow-up. This patient had no significant macroproactin present and the tumour demonstrated negative immunohistochemical staining for proactin. Preoperative imaging demonstrated an empty sella.

Preoperative hormonal data for incidentalomas

Those with incidentalomas had fewer individual hormone deficiencies compared to those presenting with symptoms reaching significance for nil deficiency, gonadal and adrenal axes (Fig. 2a). The prevalence of those with multiple hormone deficiency was not significantly different to those presenting symptomatically: 28% vs 43%, respectively, $P = 0.164$.

### Table 1. Variables associated with multiple hormone deficiency (MHD) pre- and postoperatively

<table>
<thead>
<tr>
<th>Variable</th>
<th>Preoperative</th>
<th>P-value</th>
<th>Postoperative</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gender subgroup (%)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>20/60 (67)</td>
<td>0.038</td>
<td>22/48 (46)</td>
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<td>10/35 (29)</td>
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<td>10/28 (36)</td>
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<td>6/22 (27)</td>
<td>0.15</td>
<td>2/14 (14)</td>
<td>0.15</td>
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<tr>
<td>Gender subgroup (%)</td>
<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>Male equivalent age</td>
<td>8/19 (42)</td>
<td>0.32</td>
<td>6/16 (38)</td>
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</tr>
<tr>
<td>Premenopausal female</td>
<td>6/22 (27)</td>
<td>0.15</td>
<td>2/14 (14)</td>
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<tr>
<td>Age (years)</td>
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<tr>
<td>MHD present</td>
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<td>0.37</td>
<td>60.6 ± 2.1</td>
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<tr>
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<td>56.4 ± 1.7</td>
<td>0.24</td>
<td>54.8 ± 2.1</td>
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<tr>
<td>Size (mm)</td>
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<tr>
<td>MHD present</td>
<td>30.5 ± 1.6</td>
<td>0.31</td>
<td>33.0 ± 1.9</td>
<td>0.47</td>
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<tr>
<td>Not present</td>
<td>28.3 ± 1.3</td>
<td>0.31</td>
<td>28.3 ± 1.4</td>
<td>0.47</td>
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<tr>
<td>Invasion (%)</td>
<td></td>
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<tr>
<td>Yes</td>
<td>20/54 (37)</td>
<td>0.84</td>
<td>14/32 (44)</td>
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<td>No</td>
<td>20/55 (36)</td>
<td>0.84</td>
<td>28/53 (52)</td>
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<td>Incidental (%)</td>
<td></td>
<td></td>
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</tr>
<tr>
<td>Yes</td>
<td>11/40 (28)</td>
<td>0.104</td>
<td>9/31 (29)</td>
<td>0.22</td>
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<td>No</td>
<td>33/77 (43)</td>
<td>0.72</td>
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<td>Multivariate analysis</td>
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<tr>
<td>Gender</td>
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<td>OR</td>
<td></td>
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</tr>
<tr>
<td>(male vs female)</td>
<td>1.08</td>
<td>1.08</td>
<td>1.08 (1.00)</td>
<td>0.99</td>
</tr>
<tr>
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<td>1/5 (0.20)</td>
<td>0.06</td>
</tr>
<tr>
<td>Size</td>
<td>1.03±0.03</td>
<td>0.25</td>
<td>1.00±0.04</td>
<td>0.04</td>
</tr>
</tbody>
</table>

OR, odds ratio.

*Male vs females.

$P$Premenopausal females vs others.

Postoperative hormonal data

Hormonal data 6 months after surgical treatment were available for 94 cases: 51 men, 29 postmenopausal women and 14 premenopausal women (Fig. 1b). Individual hormone deficiencies of gonadal, thyroid and adrenal axes were seen less frequently in premenopausal women, but reached significance only for gonadal deficiency compared to men. Multiple hormone deficiency occurred more frequently in men 22/48 (46%) and postmenopausal women 10/28 (36%), compared with premenopausal women 2/14 (14%) ($P = 0.049$).

Univariate analysis for the presence of postoperative multiple hormone deficiency just failed to be associated with age at presentation ($P = 0.055$) but was associated with larger tumour size ($P = 0.045$) (Table 1). When compared to men of the same age, premenopausal women were less likely to have multiple hormone deficiency but this did not reach significance: 38% vs 14%, respectively, $P = 0.15$. When multivariate analysis for mal-

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Gender differences in presentation and outcome of nonfunctioning pituitary macroadenomas

Fig. 2 (a) Preoperative and (b) postoperative hormone deficiency in incidentalomas and symptomatic cases. Incidental vs symptomatic cases respectively. (a) Nil (95.9%) vs 14/79 (18%), *P < 0.005: Gonadal axis 19/39 (47%) vs 38/76 (63%) , †P < 0.005: Thyroid axis 13/40 (33%) vs 22/77 (29%), P = 0.04; Adrenal axis 4/41 (10%) vs 24/78 (31%), †P = 0.01; Multiple 11/40 (28%) vs 33/77 (43%), P = 0.06; (b) Nil 10/31 (32%) vs 17/57 (30%), P = 0.8; Gonadal axis 14/30 (47%) vs 25/55 (46%), P = 0.13; Thyroid axis 12/32 (38%) vs 20/58 (34%), P = 0.78; Adrenal axis 3/33 (24%) vs 11/59 (18%), P = 0.26; Multiple 9/31 (29%) vs 25/59 (42%), P = 0.22.

Table 2. Postoperative: new hormonal losses by gender subgroups. Ninety-three cases pre- and postoperative paired data (14 premenopausal females, 29 postmenopausal females and 50 males)

<table>
<thead>
<tr>
<th></th>
<th>Premenopausal females (%)</th>
<th>Postmenopausal females (%)</th>
<th>Males (%)</th>
<th>Total (%)</th>
<th>P-value Females vs males</th>
<th>P-value Premenopausal females vs others</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gonadal axis loss</td>
<td>1/4 (25)</td>
<td>4/11 (36)</td>
<td>4/14 (28)</td>
<td>9/29 (31)</td>
<td>0.78</td>
<td>0.78</td>
</tr>
<tr>
<td>Thyroid axis loss</td>
<td>1/9 (11)</td>
<td>2/18 (11)</td>
<td>7/27 (26)</td>
<td>9/54 (17)</td>
<td>0.16</td>
<td>0.53</td>
</tr>
<tr>
<td>Adrenal axis loss</td>
<td>1/10 (10)</td>
<td>4/24 (17)</td>
<td>14/38 (37)</td>
<td>19/72 (26)</td>
<td>0.035</td>
<td>0.2</td>
</tr>
</tbody>
</table>

Table 3. Postoperative: hormonal recovery by gender subgroups

<table>
<thead>
<tr>
<th></th>
<th>Premenopausal females (%)</th>
<th>Postmenopausal females (%)</th>
<th>Males (%)</th>
<th>Total (%)</th>
<th>P-value Females vs males</th>
<th>P-value Premenopausal females vs others</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gonadal axis recovery</td>
<td>6/10 (60)</td>
<td>3/12 (25)</td>
<td>5/16 (31)</td>
<td>14/53 (26)</td>
<td>0.04</td>
<td>0.007</td>
</tr>
<tr>
<td>Thyroid axis recovery</td>
<td>3/4 (75)</td>
<td>2/9 (22)</td>
<td>9/23 (39)</td>
<td>14/36 (39)</td>
<td>0.97</td>
<td>0.23</td>
</tr>
<tr>
<td>Adrenal axis recovery</td>
<td>5/10 (100)</td>
<td>0/5 (0)</td>
<td>5/10 (50)</td>
<td>0/10 (0)</td>
<td>0.6</td>
<td>0.03</td>
</tr>
</tbody>
</table>

Postmenopausal female vs others.
†P = 0.02.

New hormonal deficiency postoperatively

Paired pre- and postoperative hormones were available for 93 cases: 50 men, 29 postmenopausal women and 14 premenopausal women. A new hormonal deficiency occurred in 17 (31%) of cases. Men were more likely than women to develop secondary adrenal insufficiency postoperatively (37% vs 15%, P = 0.033) (Table 2). A new classification of multiple hormone deficiency was observed in 25% of cases.

Hormonal recovery postoperatively

Hormonal recovery was observed overall in 26. 62% of cases and 39% classified as having multiple hormone deficiency prospectively recovered at least one hormone axis to have ≤ 1 hormone deficiency. The greatest recovery occurred in premenopausal women (60% gonadal, 75% thyroid, 100% adrenal), reaching significance for the gonadal and adrenal axes (Table 3).

Discussion

Nonfunctioning pituitary macroadenomas are associated with hormone deficiencies pre- and postoperatively, occurring with greater frequency compared with secretory adenomas and when NEPMAs are resected transcranially compared with the
trans-sphenoidal route.\textsuperscript{1,4,12} Data regarding hormone deficiencies have generally been reported with genders combined, except for occasional differentiation when describing hypogonadism. This report suggests that there may be gender differences in presentation and postoperative outcomes, which relate to tumour size and possibly also by disease duration. Men with NFPMA appear to have a higher prevalence of multiple pituitary hormone deficiency at presentation, while premenopausal women have a more favourable hormonal outcome following transsphenoidal pituitary surgery.

At presentation each gender subgroup had similar frequency of single hormone deficiencies in the order of gonadotrophin > thyroid hormone > pituitary adrenal axis. Secondary hypogonadism occurred in two-thirds of cases with no difference between the groups. Five small series have reported on the prevalence of pituitary dysfunction in NFPMA according to gender with reference to hypogonadism occurring in 69% men and 60%100% in women.\textsuperscript{5,11,13} Gender-specific deficiencies of other hormonal axes have not been reported.\textsuperscript{9,11,18}

We have observed a high prevalence of incidentally discovered NFPMA occurring in one-third of the cohort compared with 18% in other studies.\textsuperscript{14,15,17,21,26,27} Our series which spans a more recent time period likely represents the widespread availability of neuroimaging. These incidentalomas were prevalent in older age, confounded by a higher prevalence in postmenopausal women (as this was not observed in men). This gender difference has not been previously reported and may relate to postmenopausal women being asymptomatic of gonadial deficiency, thus prolonging presentation to medical attention. One small study observed that 22 elderly patients (>70 years) matched with younger controls with similar type and size of pituitary adenoma were less likely to present with symptomatic hormone deficiency despite having the same number of deficiencies biochemically.\textsuperscript{7,28} In our series, incidentalomas had a similar rate of multiple hormone deficiency to those presenting symptomatically, although they did have a lower frequency of individual hormone deficiencies.

Serum prolactin was commonly above gender-specific reference ranges, ranging from 40% in men to 68% in women; these findings are higher than described previously, probably as gender-specific references ranges were used here which was not distinguished in most other series.\textsuperscript{6,11,15,17,20,22,29} Prolactin levels were above 2000 mU/L in only four cases, supporting the concept that disconnection hyperprolactinaemia in NFPMA produces levels almost always below 2000 mU/L.\textsuperscript{30}

Postoperatively, premenopausal women had the lowest hormone deficiencies for each individual hormonal axis, reaching significance for the gonadal axis, and were less likely to have multiple hormone deficiency.

There are several interesting findings from these data. Firstly, at presentation, men appear more likely to have multiple hormone deficiency compared with women. Men did tend to have larger adenomas than premenopausal women (and significantly to those of comparable age) but not postmenopausal women. Premenopausal women may have smaller adenomas as a result of presenting sooner in the course of the disease caused by menstrual abnormalities. Overall, however, size and invasiveness were not useful in predicting hormonal deficiencies at presentation, consistent with other series.\textsuperscript{18,31}

Postoperative multiple hormone deficiency, however, was influenced by tumour size. Premenopausal women appeared to have less individual and multiple hormone deficiencies compared with men, (including those of comparable age). Although not reaching significance for all axes, this is likely due to a type 2 error resulting from the small sample size. Recovery of gonadotrophin secretion is of no clinical benefit to postmenopausal women but is a major issue for women of reproductive age with the importance of potential fertility.

If tumour size alone determined postoperative hormonal outcome, then incidentalomas should also have fewer multiple hormone deficiency postoperatively as these tumours were significantly smaller than those presenting symptomatically and were of similar size to premenopausal women. However, this study did not find improved hormonal outcomes from incidentalomas. Incidentalomas occurred in those much older than those presenting symptomatically. Although age just failed to be associated with multiple hormone deficiency postoperatively, it is possible that increasing age plays a limiting role in the capacity for hormonal recovery. It may be that disease duration rather than chronological age is more influential for the ability of hormonal restoration.

Given tumour size and possibly shorter disease duration appear to influence postoperative hormone status, these factors may relate to differences in intrasellar pressure (ISP). It has been postulated that higher ISP reduces portal blood flow to the normal pituitary, risking necrosis of viable pituitary cells. Three studies have investigated ISP in pituitary adenomas at presentation and one study postoperatively in apoplectic cases. Two of the three series demonstrated ISP was higher in those with hypopituitarism at presentation,\textsuperscript{11,13} while one study did not.\textsuperscript{31} These three studies did not show a relationship between ISP and adenoma size, while neither gender differences nor the influence of ISP on postoperative hormonal recovery were assessed.\textsuperscript{31,32,33} The fourth study demonstrated less postoperative hypopituitarism in those with lower ISP in cases undergoing surgery for apoplexy.\textsuperscript{24} Thus, it may be possible that premenopausal women with smaller tumours and of shorter disease duration may have lower ISP, resulting in less damage to the normal pituitary cells and thus improved restoration of pituitary function postoperatively.

Several limitations need to be considered in this retrospective observational study. Firstly, GH status has not been reported as few data were available in our series. GH deficiency is reported in some series, and when reported, is the commonest axis deficiency ranging from 61.5% to 100%.\textsuperscript{6,11,14,17,29} In Australia, adult GH replacement is not funded either by the government or by private health insurance, resulting few who are able to self-fund. Consequently, the assessment of growth hormone deficiency has not been routine practice postpituitary surgery generally reserved for those with financial means to afford replacement. Secondly, it is difficult to make direct comparisons of hormonal data between other studies in this field as the literature is heterogeneous regarding study size, definition of hypopituitarism, inclusion of apoplectic and other pituitary adenomas (micro and functioning.
adenomas), and in older studies including cases with craniomegaly. We have chosen to consider multiple hormone deficiency (≥2 axes deficient) as clinically distinct from one or no hormone deficiency as this demonstrates greater pituitary pathological damage and of clinical relevance to practice.

Thirdly, elegant studies in the past have undertaken detailed dynamic testing of pituitary function. Few patients had a dynamic test preoperatively, and some patients who were considered cortisolsufficient preoperatively may in fact not have passed such an assessment; however, the finding of a quarter of combined cohort having this deficiency at presentation is compatible with other series. The diagnosis of pituitary adenoma axis deficiency postoperatively was based initially on repeated postoperative morning cortisol levels, with dynamic testing as indicated.

Finally, missing follow-up data from patients reflect the tertiary neurosurgical practice, with postoperative follow-up and assessment being carried out by the referring endocrinologist from other centres or private practice in approximately 20% of cases. This degree of missing follow-up data is comparable with other retrospective studies in this field.

In summary, this series of nonfunctioning pituitary macroadenomas operated on in an Australian tertiary referral centre shows that postoperatively premenopausal women tend to have fewer deficiencies and an increased likelihood of gonadal function restoration. This may relate to smaller tumour size associated with a shorter duration of disease process. There should be no hesitation in offering pituitary surgery to premenopausal women with nonfunctioning macroadenomas, who have the most to gain in terms of restoration of hormonal function. The novel finding of this study showing that premenopausal women have favourable hormonal outcomes will be confirmed by a prospective study.

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Competing interests/financial disclosure
Nothing to declare.

References

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4.3 Summary and conclusion

In this first description of the largest Australian cohort of operated cases of NFPMAs from a single tertiary centre between 1995-2010, it was found that hormonal differences exist between males and females which may relate to adenoma size.

This study found that males have a higher prevalence of multiple hormone deficiencies than females at presentation, although the order of hormone deficiencies was similar with gonadotropin deficiency being the most frequent axis affected in both genders. Post-operatively, males again had a higher prevalence of multiple hormone deficiencies than females, but this reached significance only compared to pre-menopausal females. Pre-menopausal females had excellent hormonal outcomes with very few hormone deficiencies post-operatively and were more likely to restore gonadal function (which occurred infrequently in males and post-menopausal females). Adenoma size was found to be smaller in pre-menopausal females. In a multivariate analysis for having post-operative multiple hormone deficiencies adenoma size was identified as the significant associated factor.

These findings have a major impact on the management of NFPMAs. Firstly, that understanding that pre-menopausal females having surgery for NFPMAs have the best hormonal outcomes has major implications on fertility potential; this is a clinical consideration for the multidisciplinary team when considering the optimal time for surgical intervention in pre-menopausal females. Secondly, these findings assist in identifying expected hormonal deficiencies in patients with NFPMAs at presentation and follow-up, with a high suspicion of males having multiple hormone deficiencies compared to females; this is of clinical importance for communicating to patients and managing patient expectations post-surgery.

The findings of this study set the foundations for the work described in the next chapter.

Having identified adenoma size as a factor associated with hormone deficiencies, I wanted
to explore another less well defined factor of intrasellar pressure (ISP) and its potential relationship between adenoma size and hormone deficiencies in NFPMAs.
Chapter 5: The relationship between intrasellar pressure, adenoma size and hormonal outcomes in operated cases of non-functioning pituitary macroadenomas

5.1 Introduction

In the previous chapter of this thesis (Chapter 4), I have described the novel findings of differences in the frequency of pituitary hormone deficiencies between genders at presentation and post-operatively in non-functioning pituitary macroadenomas (NFPMA). Pre-menopausal females had particularly favourable outcomes post-surgery with very few having any hormone deficiencies; additionally, they had a greater propensity to restore hormone function post-operatively compared to males and post-menopausal females (201). In a multivariate analysis of factors associated with post-operative multiple (≥2) hormone deficiencies (MHD) larger adenoma size remained the significant factor. An additional novel finding in this study was that pre-menopausal females were found to have smaller adenomas than males (and in males of the same age). My proposed theory to explain this favourable hormonal outcome in pre-menopausal females with NFPMA is a complex interplay between smaller adenoma size and shorter disease duration, and possibly lower intrasellar pressure (ISP).

As addressed in the literature review in Chapter 1, the relationship of ISP to adenoma size and hormone function in NFPMAs remains to be defined given the limited information on the subject. Higher ISP in pituitary adenomas is postulated to reduce portal blood flow to the pituitary, leading to impairment of hormonal signalling from the hypothalamus, reduced nutrient delivery to the normal pituitary and hence, necrosis of viable pituitary cells (113). The relationship between hormone function and ISP has been studied with particular interest by two author groups, both of whom have found that higher ISP is associated with more hormone deficiencies at presentation (113) (185). But in relation to adenoma size, studies of ISP have been mostly concordant with ISP not being related to adenoma size.
Studies have reported ISP in empty sella syndrome, non-visible adenomas and microadenomas ranging between 7-19mmHg (185) (188): while in macroadenomas ISP has been found to be much higher ranging between 20-33mmHg (184) (185) (187) (188).

Interestingly, studies so far have not found that ISP is related to adenoma size in macroadenomas. However, studies have found that once pituitary adenomas become invasive (breaching the confines of the sellar space) then ISP is generally lower than adenomas confined to the pituitary sella.

Having highlighted the importance of adenoma size as a significant factor in hormonal deficiencies in Chapter 4 and a small body of literature suggesting higher ISP is associated with more hormone deficiencies, it is imperative to attempt to investigate what relationships may exist between ISP, adenoma size and hormonal outcomes in NFPMAs. Clarifying the above relationships should lead to a deeper understanding of pathophysiology of hormone deficiencies in NFPMAs. Importantly, such knowledge is likely to be of clinical relevance when managing these cases.
5.2 Hypotheses

In the literature the relationship between ISP, adenoma size and hormone function in NFPMAs remains to be fully defined. I have previously demonstrated that in NFPMAs undergoing surgery that pre-menopausal females have smaller adenomas and they have fewer hormone deficiencies post-operatively (201). Based on this, it is hypothesised that:

1) ISP is correlated with adenoma size in NFPMAs with lower ISP occurring in smaller macroadenomas;

2) ISP is correlated with pituitary hypofunction either pre- or post-operatively with higher ISP occurring in those with hormonal axis deficiencies.
5.3 Aims

This study has been designed to explore ISP and determine if relationships exist between ISP, adenoma size and hormone function in NFPMAs undergoing surgery.

Primary Aims

3) To determine if there is a relationship between ISP and adenoma size in NFPMAs;
4) To determine if there is a relationship between ISP and pre- and post-operative hormone deficiencies in NFPMAs.

Secondary Aims

1. To determine if ISP is correlated with pre- and post-operative prolactin levels in NFPMAs;
2. To determine if ISP differs between genders in NFPMAs;
3. To compare ISP in NFPMAs to functioning pituitary adenomas.
5.4 Methodology

Consecutive participants with NFPMAs or functioning pituitary adenomas undergoing primary pituitary surgery were prospectively recruited from St Vincent’s Public and Private Hospitals, Melbourne, Victoria, Australia, between September 2015 and December 2018. Written informed consent was obtained from all cases and this study was approved by the local ethics committee.

Adenoma size was measured by pre-operative gadolinium contrast Magnetic Resonance Imaging (MRI) measured in three dimensions (height, width, depth). Size was classified by the following: microadenoma (<10mm) or macroadenoma (≥10mm), maximum diameter and volume (Di Chiro and Nelson method π/6 (a.b.c)). Non-visible adenomas were those that could not be appreciated on MRI but were confirmed as adenomas on histological examination post-operatively. Site specific location of adenomas was defined as non-invasive if lesions were intra or suprasellar and invasive if having perforated into the sphenoid sinus, having cavernous sinus extension (>180 degrees circumferential carotid artery) or having perforated the diaphragma sellae as determined by pre-operative MRI findings.

Pre-operative blood tests for static pituitary hormones of TSH, T4, early morning cortisol, FSH/ LH, testosterone, oestradiol, growth hormone (GH), IGF-1 and prolactin were collected from all cases. Post-operative hormone assessment for cases with NFPMAs was determined at six months as this is when hormone deficiencies are known to be well established (202). Blood tests were undertaken by numerous pathology services (as is the usual standard of care in Australia, with multiple services available). Hormone deficiency for each axis was defined as results below the reference range for T4 with inappropriately low or normal TSH, testosterone and oestradiol (with inappropriately low gonadotropins), or the requirement for hormone replacement. Where clinically indicated, the insulin tolerance test (ITT), glucagon stress test, and short Synacthen tests were used for diagnosing GH and cortisol deficiencies. GH deficiency was the one hormone axis that has not been reported in this
study (nor in the previous study in Chapter 4). Although GH deficiency is the commonest hormone deficiency in NFPMA, diagnosis is difficult as it requires either a stimulatory test (which can only be done at selected tertiary centres) or by evidence of at least three hormone axis deficiencies and low IGF-1 (203). Historically in Australia, few cases have undergone stimulatory tests for diagnosing GH deficiency post-operatively, as before 1st December 2018, GH treatment was not listed on the Pharmaceutical Benefits Scheme and consequently was not routinely prescribed due to prohibitive costs to patients. Therefore, for the 41 cases of NFPMAs in this current study, no cases underwent a stimulatory test for the detection of GH deficiency pre- or post-operatively.

ISP was measured at the time of surgery using a commercial kit - CODMAN® MICROSENSOR™ ICP Transducer (Camino Laboratories San Diego CA) (113) (187) (188). At surgery, once the case was anaesthetised, the dura was initially exposed by removal of the bony sellar floor. This approach is part of the normal surgery for the removal of pituitary adenomas. Instead of initially fully opening the dura to access the pituitary adenoma, a 1.5mm dural opening was made allowing insertion of the fibreoptic transducer tip into the pituitary fossa to reduce the chances of adenoma extravasation (Figure 5.0 A and B). When a stable waveform was present on the pressure monitor, ISP was recorded at baseline with measurement of arterial carbon dioxide. ISP was measured under normal physiological conditions of normocapnia (PaCO2 35-45mmHg) at the commencement of surgery. After initial ISP was taken the patient was hyperventilated to reduce arterial carbon dioxide levels. This was done to assess the effect of hypocapnia on ISP. Most studies of ISP have been done under normocapnic conditions, but two studies have reported ISP in hypocapnia conditions (186) (194). The results of inducing hyperventilation in this study showed that ISP decreased by a mean of 3.08 ± 0.7mmHg (95% CI 1.8-4.3) (p=<0.01) from initial measurement to the final post-hyperventilation measurement. Thus, the ISP measurement used in this study was the initial measurement done prior to hyperventilation. The measurement of ISP was undertaken by two specialist pituitary surgeons throughout the study (PMN and YYW), with prior experience of using the CODMAN® MICROSENSOR™ ICP Transducers (Figure 5.0.1 A and B).
Figure 5.0 A and B. Measuring ISP at the time of operation.

A: Surgeon PMN in operating theatre with a case undergoing ISP measurement. Red arrow points to ISP transducer probe in pituitary adenoma through patient’s nose. Blue arrow indicates the monitor for recording ISP.

B: Demonstrates the ISP probe inserted through a small opening in the dura into the pituitary adenoma (blue arrow).
In some cases, adenoma extravasation occurred through the dural opening and ISP measurements could not be undertaken. This situation was more likely to occur in apoplexy (due to the haemorrhage present) and in cystic pituitary adenomas (when the cyst ruptures). The figure below demonstrates a case of a large cystic non-functioning pituitary adenoma where the adenoma began extravasating at the time of dural opening, and ISP measurement was not able to be recorded (Figure 5.1 A and B).

Figure 5.1 A and B. An example of a cystic NFPMA in a case where ISP could not be measured due to adenoma extravasation.

A: MRI coronal view. Blue arrow demonstrates the cystic component of the adenoma.

B: MRI sagittal view. Blue arrow demonstrates the cystic component of the adenoma.
5.5 Statistical analysis

Data were explored with all cases of NFPMAs included and then with two cases of apoplexy excluded. This was done as apoplexy is a unique situation where extremely high ISP has been reported and is associated with a higher prevalence of hormone deficiencies (10) (187). As there were only two cases of apoplexy, the exclusion of these cases did not have a substantial impact on the results. Data are presented as percentages for categorical variables and mean ± standard deviation (SD) for continuous variables. Age was analysed by tertiles. ISP and adenoma volume data are skewed to the right so are presented as median and interquartile range (IQR). Mann-Whitney and Kruskal-Wallis tests were utilised for comparing medians between two and three or more groups, respectively. Cuzick’s test for trend was used for analysing median ISP and adenoma volumes across ordered groups for the number of hormone deficiencies. Linear regression was used to examine relationships between continuous variables of ISP, age and adenoma size volume. ISP and adenoma volume were also converted to a logarithmic scale in order to perform regression analyses.

Differences in frequency of categorical variables of adenoma type, the number of hormone deficiencies (0, 1 or MHD) at presentation and post-operatively, and gender were analysed by chi-squared tests. Binary logistic regression was used to assess the probability of hormone deficiencies (no hormone deficiency compared to one or more hormone deficiencies) using the explanatory variables of ISP and adenoma volume (log transformed) as these were the two variables which showed evidence of a relationship to hormone deficiencies. A two-tailed p value <0.05 was deemed statistically significant. The data was analysed using statistical package Minitab 18.

In order to calculate a sample size to detect a difference in ISP between those with and without hormone deficiencies, I used ISP measurements based on the study by Arafah et al, whereby mean ISP was 19.9 mmHg (SD 7.3) in those without pre-operative hormone deficiencies, and 33.6mmHg (SD 13.3) in those with one or more hormone deficiency (p<0.005) (113). Assuming similar means and variability, to compare a difference in ISP
between groups with no hormone deficiencies versus one or more hormone deficiencies (with parameters set at $\alpha = 0.05$, power of 0.8), then 9 cases for each group would be required, in order to show a significant difference. From the retrospective cohort from Chapter 4, approximately a third of cases had no hormone deficiency at presentation and the remainder had one or more hormone deficiencies. I aimed to recruit 40 cases of NFPMA to ensure approximately one third (n=12) of this group would have no hormone deficiencies.

This study aimed to recruit half the number of cases of functioning pituitary adenomas (n=20), which was estimated to be a sufficient number to use as a comparator for the cases of NFPMAs.
5.6 Results

5.6.1 Demographic results

In total, 41 cases with NFPMAs were recruited and are included in the demographic and hormonal analyses. Of these, 24 were males and 17 were females (12 post-menopausal females and five pre-menopausal females), with mean age of 60.6 (12.3) years (range 27-78) (Table 5.0).

Of the 41 cases, 21 (51.2 %) presented symptomatically with mass effect (headache or visual changes) or hormonal complaints, 17 (41.5%) presented incidentally and three cases presented due to acute apoplexy (7.3%).

All cases with NFPMAs were macroadenomas and 10/41 (24.4%) of cases had evidence of invasion at presentation (Table 5.0).

Of the 41 cases recruited ISP data was available in 35 cases. Six cases were excluded due to two having adenoma extravasation (one of these cases had pituitary apoplexy and the other case has a cystic adenoma) and four because of ISP equipment issues (two cases due to the ISP probe not being available in the operating room, one case where the ISP probe failed to take a measurement and one cases due to misplaced ISP data). Of the 35 included cases with ISP data three cases had cystic adenomas that did not extravasate and there were two cases of acute pituitary apoplexy.

Pre-operative hormonal status was recorded in all 41 cases of which 14 (34.1%) had no hormone deficiency, 12 (29.3%) had one hormone deficiency and 15 (36.6%) had MHD (Table 5.0). Males appeared to have more MHD than females (41.7% vs 23.5%) although this difference was not statistically significant.
Post-operative hormonal outcomes were recorded in 40 cases of which 19 (47.5%) had no hormone deficiency, 9 (22.5%) had one hormone deficiency and 12 (30%) had MHD (Table 5.0). MHD was more common in males compared to females (39.1% vs 17.6%), although again, this difference was not statistically significant. None of the five pre-menopausal females in this study had post-operative MHD (Table 5.0).

The order of hormone axis deficiency at presentation was 24/41 (58.5%) for gonadotropin, 10/41 (24.4%) for TSH and 11/41 (26.8%) for the ACTH-cortisol axis. The order of hormone axis deficiency post-operatively was 20/40 (50%) for gonadotropin, 11/40 (25.7%) for TSH and 4/40 (10%) for the ACTH-cortisol axis.

Males tended to have larger adenomas than females as measured by maximum adenoma diameter (p=0.06) and by volumetric analysis (p=0.024). This difference was mainly accounted for by smaller adenoma size in the pre-menopausal females (Table 5.0).

5.6.2 Results of primary and secondary end points of ISP in NFPMA

Primary end points of ISP in NFPMA

Median ISP (IQR) in the cohort was 22mmHg (16, 29) (range 7-78) and was similar even after the two cases of pituitary apoplexy were excluded from the data (Table 5.1). In the two cases of apoplexy, ISP was 19mmHg in a 27 year old male and markedly elevated at 51mmHg in a 45 year old pre-menopausal female. The highest ISP recorded in this series occurred in the case of a 65 year old male with an ISP of 78mmHg. Repeated sampling during hyperventilation produced similar results signifying the validity of this high reading.
ISP was considered separately in cases of NFPMAs by location of non-invasive adenomas (sella/ suprasellar) and invasive adenomas. In nine cases with invasive adenomas median ISP was 19mmHg (15.5, 24.5) and appeared somewhat lower compared to 29 cases with non-invasive adenomas with median ISP of 22mmHg (15.5, 31), although this difference in ISP was not statistically significant (p=0.57) (Table 5.2). It was also noted that cases with invasive disease had significantly larger adenoma volumes compared with non-invasive adenomas (p=0.005) (Table 5.2).

There was no significant relationship found between ISP and adenoma size, measured by maximal diameter or by volume, when these were analysed by linear regression analysis (p=0.67 and p=0.49, respectively) (Table 5.1 and Figure 5.2 A). The relationship of ISP and adenoma size was also analysed by considering groups of invasive and non-invasive NFPMAs as mentioned above. Although linear regression did not find a significant relationship between ISP and adenoma volume, it was noted that the slopes of the regression lines were in opposite directions for each group (Table 5.2 and Figure 5.3 A and B).

Median ISP was found to be 20mmHg in those with no hormone deficiency, 20.5mmHg in those with one hormone deficiency and 28.5mmHg in those with MHD pre-operatively (trend analysis, p=0.038): similar results were obtained when the two cases of apoplexy were excluded from the analysis (Table 5.1 and Figure 5.4). When groups were examined by either no pre-operative hormone deficiency or any hormone deficiency ISP was found to be 20mmHg (10, 24) in those with no hormone deficiency and 23mmHg (16.5, 34.8) in those with one or more hormone deficiency: higher ISP appeared to be associated with having any hormone deficiency, although this difference did not reach statistical significance, p=0.088, (Table 1.1 and Figure 5.5). Logistic regression analysis was used to examine the effect of ISP on the probability of having a pre-operative hormone deficiency. For each doubling of ISP, the estimated odds of developing a hormone deficiency increased by a factor of 3.3 (95% CI 0.97-11.4), p=0.055; a relationship that just failed to reach statistical significance.
Post-operatively, median ISP was found to be 20mmHg in those with no hormone deficiency, 23mmHg in those with one hormone deficiency and 24.5mmHg in those with MHD post-operatively; similar results were obtained when the two cases of pituitary apoplexy were also excluded from the analysis (see Table 5.1). When groups were examined by either no post-operative hormone deficiency or any hormone deficiency ISP was found to be 20mmHg (14, 25.8) in those with no hormone deficiency and 22mmHg (16, 34) in those with one or more hormone deficiency: no significant difference in median ISP was found between these two groups, p=0.37 (Table 5.1).

*Secondary end points of ISP in NFPMAs*

ISP was investigated by gender groups. Of the 35 cases with ISP data, there were 20 males and 15 females (12 post-menopausal females and 3 pre-menopausal females) (Table 5.1). Excluding the two cases of apoplexy, median ISP was 22.0mmHg (16-30) in males and 21.0mmHg (10-24) in females, and this difference was not statistically significant (p=0.13). In the two pre-menopausal females in the cohort (one excluded due to apoplexy) ISP was 7mmHg and 22mmHg (median ISP of 14.5mmHg) (Table 5.1 and Figure 5.6).

Regarding the relationship of ISP to pre- and post-operative prolactin levels, there was no correlation between the two variables (Table 5.1).

ISP in NFPMAs was compared to ISP in 18 cases of functioning adenomas. In these 18 cases of functioning adenoma median ISP was 22.5mmHg (14.5, 32) (range 8-57); this value was similar to ISP in 35 cases of NFPMAs with median ISP of 22mmHg (16, 29), p=0.7. This is shown in Figure 5.7.

The cohort of 18 cases of functioning adenomas consisted of non-lesional adenomas (n=2), microadenomas (n=5) and macroadenomas (n=11). Median ISP in patients with functioning
microadenomas and macroadenomas were not significantly different (19 mmHg vs 23 mmHg, respectively, p=0.9) as shown in Figure 5.8. In the two cases of non-lesional adenomas ISP was 12 mmHg and 28 mmHg. Detailed analyses of ISP in functioning adenomas can be found in Appendix 1. There was no significant correlation found between ISP and adenoma size (maximal diameter or volume) in the functioning adenomas. When NFPMAs and functioning adenomas were combined there was still no significant correlation found between ISP and adenoma size (Appendix 1, Table 1.1).

5.6.3 Results of other analyses of ISP in NFPMAs

No significant relationship was seen between ISP and increasing age when analysed as either a categorical or a continuous variable (Table 5.1).

There was no significant difference in median ISP whether cases presented incidentally or with symptoms; or if presenting with or without headaches (Table 5.1).

The number of ISP procedures were done equally between the two operating neurosurgeons. PMN undertook 17 ISP measurements and YYW undertook 18 measurements. Median ISP was not different between surgeons (Table 5.1).

The relationship between ISP and differences of individual hormonal axis deficiency (gonadotropin, TSH, ACTH-adrenal axis) pre- and post-operatively were also examined. No significant differences in median ISP were found in the individual hormonal axis comparing those with and those without the particular axis deficiency. These are presented in Appendix 2 in Table 2.0 A and B for pre-operative axis deficiency and Table 2.1 A and B for post-operative axis deficiency.
As adenoma volume is known to be a factor associated with hormone deficiencies analyses were also undertaken in this current cohort of NFPMAs to assess the relationship of hormone deficiencies to adenoma size. Further details of these results can be found in Appendix 2.

Median adenoma volume in those with no hormone deficiency pre-operatively appeared to be smaller than those with one hormone deficiency and MHD (2420mm$^3$ vs 3935mm$^3$ and 3044mm$^3$, respectively), although trend analysis did not demonstrate statistical significance (p=0.16). Median adenoma volume in those with no hormone deficiency tended to be smaller than those with any hormone deficiency (2420mm$^3$ vs 3551mm$^3$; p=0.079) (shown in Appendix 2 Table 2.2).

Logistic regression analysis was used to examine the effect of adenoma volume on the probability of having a pre-operative hormone deficiency. For each doubling of adenoma volume, the estimated odds developing a hormone deficiency was 1.8 times higher (95% CI 0.92-3.6), p=0.086, however this relationship failed to reach statistical significance.

A multivariate analysis to examine the combined effect of ISP and adenoma volume on pre-operative hormone deficiencies was not undertaken at this point, given both these variables did not reach statistical significance on their own.

Median adenoma volume in those with no hormone deficiency post-operatively appeared to be smaller than those with one hormone deficiency and MHD (2680mm$^3$ vs 3044mm$^3$ and 4343mm$^3$, respectively), although trend analysis did not demonstrate statistical significance (p=0.15). Median adenoma volume in those with no hormone deficiency was 2693mm$^3$ and 3820mm$^3$ in those with any hormone deficiency; these two volumes were not statistically different, p=0.081 (shown in Appendix 2, Table 2.2).
Table 5.0 Demographic data of 41 cases of NFPMA

<table>
<thead>
<tr>
<th></th>
<th>All</th>
<th>Males</th>
<th>Females</th>
<th>Premenopausal females (Pre-MenF)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Gender subgroup</strong></td>
<td>41</td>
<td>24</td>
<td>17</td>
<td>5</td>
</tr>
<tr>
<td><strong>Age at presentation (years); mean (SD)</strong></td>
<td>60.6 (12.3)</td>
<td>62.5 (11)</td>
<td>57.9 (13.8)</td>
<td>39 (6.8)</td>
</tr>
<tr>
<td></td>
<td>(Range 27-78)</td>
<td>(Range 27-78)</td>
<td>(Range 27-78)</td>
<td>M v F p=0.27</td>
</tr>
<tr>
<td><strong>Presentation</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Incidental</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Symptomatic</td>
<td></td>
<td></td>
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<td></td>
</tr>
<tr>
<td>Apoplexy</td>
<td></td>
<td></td>
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<tr>
<td></td>
<td>17 (41.5%)</td>
<td>10</td>
<td>7</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td>21 (51.2%)</td>
<td>12</td>
<td>9</td>
<td>3</td>
</tr>
<tr>
<td></td>
<td>3 (7.3%)</td>
<td>2</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>Symptomatic group</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Headaches</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>10 (24.4%)</td>
<td>5</td>
<td>5</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td>9 (21.9%)</td>
<td>4</td>
<td>5</td>
<td>2</td>
</tr>
<tr>
<td></td>
<td>8 (19.5%)</td>
<td>6</td>
<td>2</td>
<td>1</td>
</tr>
<tr>
<td>Visual abnormality</td>
<td></td>
<td></td>
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<tr>
<td>Hormonal presentation</td>
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<td></td>
<td></td>
</tr>
<tr>
<td>Adenoma size; median (IQR)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Maximum size (mm)</td>
<td>22.0 (18.5, 27)</td>
<td>22.5 (20, 27)</td>
<td>20.0 (16.5, 24.5)</td>
<td>18.0 (16.5, 23)†</td>
</tr>
<tr>
<td></td>
<td>(Range 13-45)</td>
<td>(Range 27-45)</td>
<td>(Range 16-30)</td>
<td>M v F p=0.06</td>
</tr>
<tr>
<td>Volume (mm³)</td>
<td>2937 (2089-5287)</td>
<td>3711.0 (2558-6743)</td>
<td>2309.1 (1212-4265)</td>
<td>2002.8 (887-4943)‡</td>
</tr>
<tr>
<td></td>
<td>(Range 674-21,912)</td>
<td>(Range 769-714)</td>
<td>(Range 769-1265)</td>
<td>M v F p=0.024</td>
</tr>
<tr>
<td><strong>Invasive adenoma</strong></td>
<td>10 (24.4%)</td>
<td>6</td>
<td>4</td>
<td>1</td>
</tr>
<tr>
<td><strong>Hormone status</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Presentation</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0 HD</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1HD</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>MHD (≥2 HD)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>14 (34.1%)</td>
<td>6</td>
<td>8</td>
<td>3</td>
</tr>
<tr>
<td></td>
<td>12 (29.3%)</td>
<td>7</td>
<td>5</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td>15 (36.6%)</td>
<td>11 (45.8%)</td>
<td>4</td>
<td>1</td>
</tr>
<tr>
<td>Post-operatively</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>n=40</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0 HD</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1HD</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>MHD (≥2 HD)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>19 (47.5%)</td>
<td>9</td>
<td>10</td>
<td>3</td>
</tr>
<tr>
<td></td>
<td>9 (22.5%)</td>
<td>5</td>
<td>4</td>
<td>2</td>
</tr>
<tr>
<td></td>
<td>12 (30%)</td>
<td>9</td>
<td>3</td>
<td>0</td>
</tr>
</tbody>
</table>

HD: Hormone deficiency, MHD: Multiple hormone deficiency

† Males Vs Pre-MenF p=0.05; Kruskal-Wallis test for the three gender subgroups p=0.12
‡ Males vs Pre-MenF p=0.09; Kruskal-Wallis test for the three gender subgroups p=0.072
## Table 5.1 Intrasellar pressure in 35 cases of NFPMA

<table>
<thead>
<tr>
<th></th>
<th>Median ISP (IQR)</th>
<th>Median ISP (IQR) Excluding 2 cases apoplexy</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>All n=35</strong></td>
<td>22 (16, 29)</td>
<td>22 (15.5, 28.5) (range 7-78)</td>
</tr>
<tr>
<td><strong>Gender</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Males n=20</td>
<td>21.5 (16.5-29.8)</td>
<td>22 (16-30)</td>
</tr>
<tr>
<td>Females n=15</td>
<td>22 (10-24)</td>
<td>21 (10-24)</td>
</tr>
<tr>
<td>Pre-MenF n=3</td>
<td>22 (7-51)</td>
<td>14.5 (n=2)</td>
</tr>
<tr>
<td><strong>Age (years)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>27-60</td>
<td>18.5 (15.3, 31.8)</td>
<td>18.5 (13.5, 25.3)</td>
</tr>
<tr>
<td>61-67</td>
<td>20 (13.0, 30.0)</td>
<td>22 (12.3, 32.3)</td>
</tr>
<tr>
<td>67-78</td>
<td>24 (19.8, 27.8)</td>
<td>24 (17.5-27.5)</td>
</tr>
<tr>
<td><strong>Continuous Variable</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(correlation co-efficient with 95% CI)</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>0.09 (-0.3, 0.48)</td>
<td>0.2 (-2.5, 0.65)</td>
</tr>
<tr>
<td><strong>Presentation</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Symptomatic vs Incidental</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Headaches vs No Headaches</td>
<td>19 (16, 30) vs 23 (14.5, 28.8)</td>
<td>19 (16, 24) vs 23 (14.5, 29.8)</td>
</tr>
<tr>
<td></td>
<td>19 (16-24) vs 23 (14.5, 29.8)</td>
<td>20 (15.8, 26.8) vs 22 (14, 29)</td>
</tr>
<tr>
<td><strong>Adenoma Size</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(correlation co-efficient with 95% CI)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Maximum diameter</td>
<td>0.15 (-0.54, 0.83)</td>
<td>0.22 (-0.46, 0.91)</td>
</tr>
<tr>
<td>Volume</td>
<td>0.11 (-0.14, 0.35)</td>
<td>0.12 (-0.13, 0.36)</td>
</tr>
<tr>
<td><strong>Hormone status</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Presentation</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No HD</td>
<td>20 (10, 24)</td>
<td>20 (10, 24)</td>
</tr>
<tr>
<td>1 HD</td>
<td>20.5 (15.3, 27)</td>
<td>20.5 (15.3, 27)</td>
</tr>
<tr>
<td>MHD (≥2 HD)</td>
<td>28.5 (18, 39.5)</td>
<td>28.5 (17.5, 36.5)</td>
</tr>
<tr>
<td></td>
<td>† p=0.038</td>
<td>† p=0.047</td>
</tr>
<tr>
<td>No HD vs 1 or more HD</td>
<td>20 (10, 24) vs 23.0 (16.5, 34.8)</td>
<td>20 (10, 24) vs 23.0 (16,34.3)</td>
</tr>
<tr>
<td><strong>Hormone status</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Post-operatively</td>
<td></td>
<td></td>
</tr>
<tr>
<td>No HD</td>
<td>20 (14, 25.8)</td>
<td>20.5 (13, 25.8)</td>
</tr>
<tr>
<td>1 HD</td>
<td>23 (19, 34.5)</td>
<td>22 (19, 24)</td>
</tr>
<tr>
<td>MHD (≥2 HD)</td>
<td>24.5 (13.8, 33)</td>
<td>24.5 (13.8, 33)</td>
</tr>
<tr>
<td></td>
<td>† p=0.46</td>
<td>† p=0.49</td>
</tr>
<tr>
<td>No HD vs 1 or more HD</td>
<td>20 (14, 25.8) vs 22 (16, 34)</td>
<td>21 (12, 26.5) vs 22 (16, 29.8)</td>
</tr>
<tr>
<td><strong>Prolactin level</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>(correlation co-efficient with 95% CI)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pre-operative prolactin</td>
<td>0.002 (-0.02, 0.02)</td>
<td>0.005 (-0.01, 0.02)</td>
</tr>
<tr>
<td>Post-operative prolactin</td>
<td>0.01 (-0.05, 0.08)</td>
<td>0.01 (-0.05, 0.08)</td>
</tr>
<tr>
<td></td>
<td>p=0.79</td>
<td>p=0.59</td>
</tr>
<tr>
<td></td>
<td>p=0.69</td>
<td>p=0.69</td>
</tr>
</tbody>
</table>

HD: Hormone deficiency, MHD: Multiple hormone deficiency, Pre-MenF: premenopausal female

† Cuzick’s test for trend
### Table 5.2 Findings of cases of NFPMAs by adenoma location

<table>
<thead>
<tr>
<th></th>
<th>Non invasive (Sella / Suprasellar) NFPMAs n=26</th>
<th>Invasive NFPMAs n=9</th>
<th>P value for group comparison</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Adenoma Size</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Volume (mm$^3$) median (IQR)</td>
<td>2562.8 (1613, 4291)</td>
<td>6446.6 (3519, 8729)</td>
<td>p=0.005</td>
</tr>
<tr>
<td><strong>ISP (mmHg) median (IQR)</strong></td>
<td>22 (15.5, 31)</td>
<td>19 (15.5, 24.5)</td>
<td>p=0.57</td>
</tr>
<tr>
<td><strong>Relationship of volume to ISP</strong></td>
<td>Continuous variable (correlation co-efficient with 95% CI)</td>
<td>0.26 (-0.08, 0.59) p=0.13</td>
<td>-0.29 (-0.9, 0.33) p=0.3</td>
</tr>
<tr>
<td><strong>Hormone status</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Presentation No HD</td>
<td>9/26 (34.6%)</td>
<td>2/9 (22.2%)</td>
<td>p=0.7</td>
</tr>
<tr>
<td>1 or more HD</td>
<td>17/26 (65.4%)</td>
<td>7/9 (77.7%)</td>
<td></td>
</tr>
</tbody>
</table>
Figure 5.2 Plot of ISP by adenoma location

Data plot showing ISP for each case. Horizontal line representing median ISP of sella/suprasellar NFPMA (22mmHg) and ISP in those with invasive disease (19mmHg), p=0.57.
Figure 5.3 A. Relationship of adenoma volume to ISP in 35 cases of NFPMAs

Fitted line plot of ISP and adenoma volume demonstrating no significant relationship, p=0.49.

Figure 5.3 B. Relationship of adenoma volume to ISP in NFPMAs by location

Fitted line plot of ISP and adenoma volume considering non-invasive adenomas (sella/suprasellar adenomas) separately from invasive adenomas. Linear regression lines are non-significant but demonstrate different slope directions between the two groups.
**Figure 5.4 Plot of ISP by pre-operative hormone deficiency status of 0, 1 and MHD**

Data plot showing ISP in each case. Horizontal line representing median ISP of those with no hormone deficiency (20mmHg), one hormone deficiency (20.5mmHg) and MHD (28.5mmHg), test for trend, $p=0.038$.

**Figure 5.5 Plot of ISP by pre-operative hormone deficiency status of by no or any ($\geq 1$) hormone deficiency**

Data plot showing ISP in each case. Horizontal line representing median ISP of those with no hormone deficiency (20mmHg) and those with any ($\geq 1$) hormone deficiency pre-operatively (23mmHg), $p=0.079$. 
Figure 5.6 Plot of ISP by gender subgroups (apoplexy excluded)

Data plot showing ISP in each case by gender subgroups of males, postmenopausal females (Post-MenF) and pre-menopausal females (Pre-MenF). Horizontal line representing median ISP in males (22mmHg), Post-MenF (21mmHg) and Pre-MenF (14.2mmHg). Median ISP was not significantly different between males and females, \( p=0.13 \).
Data plot showing ISP in each case of NFPMA, functioning adenoma and the groups combined. Horizontal line representing median ISP in each group. No significant difference was found in median ISP between NFPMA and functioning adenomas (22 and 22.5 mmHg, respectively), p=0.7.
Figure 5.8 Plot of ISP in functioning adenomas by categorical size

Data plot of ISP in functioning adenoma by categorical size of non-lesional, microadenomas and macroadenomas. Horizontal line representing median ISP in each group. In two cases of non-lesional adenomas ISP was 12 and 28 mmHg (median 20 mmHg), in microadenomas median ISP of 19mmHg and macroadenomas median ISP of 23mmHg. No significant differences between median ISP were found between the groups (p=0.9).
5.7 Discussion

This study aimed to explore relationships that may exist between ISP, adenoma size and hormone function in order to further understand the pathophysiology of hormonal deficiencies in NFPMAs undergoing surgery. It is also the first study to specifically describe ISP in detail in a cohort with NFPMAs.

In 35 cases of NFPMAs median ISP was found to be 22mmHg (IQR 16-29). In the only two studies that specified adenoma type mean ISP in 8 cases of NFPMAs was 28.6 ± 12.6 mmHg (184), and in 18 cases of NFPMAs ISP was reported to be not different to their mixed cohort with mean ISP of 18.7± 10.8mmHg (188). The ISP findings in the larger cohort of NFPMAs in this thesis are somewhat similar to these study findings. The findings of median ISP in NFPMAs in this current study are also comparable to what has been described in mixed types of macroadenomas with reported means ranging between 20-33mmHg (184) (185) (187) (188). The highest ISP reported in the literature to date is 67.1mmHg and this seems comparable to the one outlier case in this current study who was found to have an ISP of 78mmHg (189).

5.7.1 ISP and adenoma size

The first of the primary aims of my study was to explore the relationship between ISP and adenoma size. This study was unable to find a relationship of ISP to adenoma size in cases with NFPMAs (either by maximal diameter or volume, and even when the cohort of NFPMAs were combined with cases with functioning adenomas - Appendix 1, Table 1.1), a finding that is consistent with what has been reported in the literature to date (113) (187) (188) (189). However, a relationship between ISP and adenoma volume exclusively in NFPMAs cannot be fully excluded at this stage as this study contained few pre-menopausal females who are known to have smaller NFPMAs, as shown in my previous study in Chapter 4 (201). The lack of pre-menopausal females in this current study skews the data towards larger adenoma volumes. The lack of a relationship between ISP and adenoma size is surprising
and does seem counterintuitive; biologically it would be expected that as an adenoma grows within the enclosed space of pituitary fossa then ISP would increase similarly. When ISP increases perfusion pressure to the normal pituitary gland must adapt and exceed ISP in order to maintain viable pituitary tissue and normal pituitary hormone function. In trying to understand a reason for the lack of relationship between ISP and adenoma size I theorise that initially at least, there may still be a relationship between a growing adenoma and ISP, but once a maximum ISP level is reached in an individual then ISP remains relatively stable irrespective of ongoing growth of the adenoma. Clearly there is no way to measure serial ISP in an individual to prove this theory as ISP can only be measured as a cross sectional data point at the time of operative intervention. Factors which determine the maximum ISP reached in an individual are however unknown. When pituitary adenomas become invasive leaving the confines of the intrasellar space ISP tends to drop. Lower ISP in invasive adenomas has been shown by others and our limited data is suggestive of this too, particularly noted by a negative regression slope seen in Figure 5.3 B (185) (188) (191).

A second finding that would support that lack of relationship between ISP and adenoma size is that even in microadenomas (which are all functioning adenomas, as non-functioning microadenomas are not operated on) ISP can still be elevated similar to that found in macroadenomas. Indeed, in this current study, median ISP was found to be just as high in the functioning microadenomas as in the macroadenomas. In two cases of non-lesional adenomas ISP was 12mmHg and 28mmHg (median 20mmHg), in eight cases of microadenomas, median ISP was 19mmHg (IQR 10.5, 44) and in macroadenomas, median ISP was 22mmHg (IQR 20-35). In fact, one case with a corticotropic microadenoma had an extremely high ISP of 52mmHg which is the highest reported ISP for a microadenoma to date. Authors Arafah et al., in eight cases of microadenomas also found ISP to be elevated with mean of 25.4mmHg (113), and authors Gondim et al., found mean ISP in 12 microadenomas to be 19.2mmHg (188), both similar values of ISP to this current study’s findings. In another study, authors Lees et al., found that in five cases of empty sella syndrome ISP was lower at 11mmHg, in seven cases of small microadenomas (0-5mm in maximal diameter) mean ISP was 12mmHg, and in larger microadenomas (6-10mm in maximal diameter) mean ISP was slightly higher at 17mmHg (185). The authors made note
of one case of a microadenoma with high ISP of 26mmHg and they expressed they were unable to explain this aberrant finding. I suspect this is indeed a true reading.

The way to confirm if ISP truly has no relationship to adenoma size in NFPMAAs is by the further recruitment of pre-menopausal females, which will give a less skewed range of adenoma size than the current cohort by including smaller macroadenomas.

5.7.2 ISP and hormonal outcomes

The next primary aim in this study was to explore the relationship of ISP to hormonal outcomes. These preliminary data do suggest that higher ISP may be associated with hormone deficiencies at presentation which is keeping with what has been described in the limited literature. Although not reaching statistical significance median ISP was found to be 20mmHg in those without hormone deficiencies at presentation and 23mmHg in those with any hormone deficiency (p=0.088). In those with MHD at presentation median ISP was higher at 28mmHg demonstrating a trend for higher ISP with more hormonal axis deficiencies. Furthermore, it was found that ISP may be able to predict the risk of a pre-operative hormone deficiency; each doubling of ISP increases the odds by 3.3 times (95% CI 0.97-11.4), p=0.055. In order to confirm these findings, recruitment of a larger cohort is required. More pre-menopausal females are also required as this group have been shown to have fewer hormone deficiencies compared to males as found in my previous study in Chapter 4 of this thesis (201).

This suggestion that higher ISP may be associated with hormone deficiencies at presentation is supported by the limited literature consisting of two studies of mixed cases of pituitary adenomas. In the first study of 49 cases by Arafah et al., these authors found lower mean ISP in those with no hormone deficiencies compared to those with one or more hormone deficiencies (19mmHg vs 33.6mmHg, p<0.005) (113). In the second study of 98 cases, lower mean ISP was found in those without any hormone deficiency (17mmHg) and highest ISP
occurred in those with 1-3 hormones deficiencies (23-24mmHg, p<0.05); these ISP findings are similar to the ISP values I found in this study. However, those with 4-5 hormone deficiencies in the study by Lees et al., did not have particularly high ISP (17mmHg) (185). Perhaps this later group had larger adenomas which were invasive explaining the lower than expected ISP value.

Higher ISP was not found to be associated with post-operative deficiencies however the small number of cases in this study with post-operative hormone deficiencies may account for this lack of association. Ongoing study with further recruitment will be able to clarify these post-operative hormonal findings.

Biologically, the mechanism of higher ISP contributing to hormone deficiencies is plausible as it may compromise normal perfusion of the pituitary gland, resulting in damage of normal pituitary tissue and thus increasing the risk of the development of hormone deficiencies. This is best exemplified in its extreme form during pituitary apoplexy where ISP has been shown to rise acutely leading to complete cessation of venous perfusion to the pituitary gland (186). Apoplexy is a rare event occurring in 0.6-9.1% of neurosurgical series of pituitary adenomas undergoing surgery and is associated with a high prevalence of hormone deficiencies due to irreversible damage to normal pituitary tissue (10). In the usual scenario of NFPMAs which do not experience apoplexy I theorise that the slow rise and/or the duration of time ISP remains elevated is possibly still compromising normal pituitary perfusion and therefore impairing normal hormone function.

ISP is likely to be only one of other factors that contributes to compromised pituitary function. Other factors such as adenoma size, disease duration and reversibility of viable pituitary tissue are also influencing factors that determine hormone deficiencies in NFPMAs. Of these factors adenoma size is the only one that is easily measurable and is an established factor contributing to hormone deficiencies in NFPMAs as shown by my findings from Chapter 4 and in the literature (102) (103) (104) (105) (106) (201). The complex interaction
between ISP, adenoma size, disease duration and viability of pituitary tissue is best illustrated by what occurs in pre-menopausal females with NFPMAs. Pre-menopausal females have fewer hormone deficiencies pre-and post-operatively, have a greater likelihood of recovering from a deficient axis and are known to have smaller adenomas than males. Pre-menopausal females do present earlier in the natural history of the disease process of NFPMAs as they present to medical attention with menstrual disturbances as a manifestation of gonadotropin deficiency which is prevalent in up to 80% (74) (107) (108) (109) (110) (111). It is then plausible that a shorter disease duration also equates to less time that raised ISP can cause irreversible damage to normal pituitary tissue.

Of relevance to the above I have noted several important differences in this current cohort compared to the historical cohort from Chapter 4 that is suggestive that this current cohort may have presented earlier in the disease process (Appendix 3, Table 3.0 for full details). These differences include the current cohort having significantly smaller and less invasive adenomas, a tendency for a higher prevalence of incidentally discovered adenomas and a higher prevalence of being eupituitary post-operatively compared to the historical cohort: despite there being no significant difference in age between the cohorts. In summary, these differences are highly suggestive that this current cohort have presented earlier in the natural history of NFPMAs. It has been recognised in the literature that surgical and hormonal outcomes are more favourable when smaller NFPMAs are operated on (204).

The contribution of ISP to predicting hormone deficiencies is difficult to define given this small cohort. A larger cohort of cases with NFPMAs will need to be studied to confirm if elevated ISP is associated with pre-operative hormone deficiencies, to conclude if there is any association with post-operative deficiencies and to be able to explore the more complex relationship to adenoma size in contributing to hormone deficiencies. Currently measuring ISP in routine practice does not appear to be helpful for predicting pre-operative hormone deficiencies as these deficiencies can already be easily diagnosed by hormonal blood tests. However, the current quest to explore and quantify how ISP contributes to the development of hormone deficiencies is paramount to the understanding of the pathophysiology of
hormone deficiencies. A better understanding of the possible role ISP may play in leading to hormone deficiencies may lead to a change in the way cases of NFPMAs are managed with consideration of surgery sooner rather than later in order to prevent new hormone deficiencies developing and in this in turn may improve patient outcomes.

5.7.3 ISP and other associations

Secondary aims of this study were to explore relationships of ISP in NFPMAs to prolactin levels, gender and compare them to functioning pituitary adenomas.

As covered earlier in the discussion median ISP in NFPMAs was not different compared to 18 cases of functioning adenomas.

This study did not find ISP to be related to prolactin levels pre- or post-operatively in NFPMAs. In the literature five studies have examined ISP and its relationship to prolactin levels in mixed type of pituitary adenomas (113) (184) (185) (191) (194) and all except one study found that higher ISP correlated with higher prolactin levels at presentation (194). It is postulated in the literature that the presence of a normal or mildly raised prolactin in NFPMAs may indicate intact pituitary gland reserve as it has been previously observed that after removal of pituitary adenomas, those with higher prolactin levels at presentation had a greater propensity of restoration of other hormonal axes (106) (109) (110) (113) (192) (193). My findings are unable to confirm this observation.

In this study median ISP values were similar between males and females. Due to insufficient numbers of pre-menopausal females in this cohort my study cannot make any conclusion regarding ISP levels in this particular gender subgroup. This later point requires further exploration because if ISP is lower in pre-menopausal females then ISP may be an important factor (in addition to smaller adenoma size) explaining their more favourable hormonal outcomes.
Although not the primary aim of this study, I also explored if ISP was associated with other parameters of NFPMAs such as age, mode of presentation (symptomatic compared to incidental), and presence of headaches in NFPMAs, but I did not find any significant relationship of these parameters to ISP.

5.7.4 ISP and apoplexy

In this study three cases of pituitary apoplexy occurred of which two were able to have ISP measurements, so only limited inference can be made about the relationship between ISP and pituitary apoplexy. Authors Zayour et al., are the only group that have reported ISP in 13 cases of apoplexy and found it to be markedly raised with a mean of 44mmHg (range 25-58) compared to their non-apoplectic series of pituitary adenomas with ISP of 33.6mmHg. In 6/13 cases of apoplexy who had persistent hypopituitarism post-operatively ISP was extremely elevated at 55.9 ± 2.4mmHg compared to the remaining seven cases who had either maintained normal or recovered hormone function with ISP of 35.9 ± 7.3mmHg (187). These authors suggested that in cases with extremely elevated ISP at the time of pituitary apoplexy ischaemic necrosis of the anterior pituitary gland occurs leading to less hormone recovery. The 2 cases in my cohort occurred in a 27 year old male and a 45 year old pre-menopausal female, with ISP of 19mmHg and 51mmHg, respectively. At presentation both had multiple hormone deficiencies which fully recovered apart from gonadotropin deficiency in the female case.

The role of ISP in apoplexy is worth exploring further as a clearer relationship may indeed exist between extremely high ISP and hormonal outcomes. If so, an ISP measurement at the time of surgery for cases of apoplexy may assist when making clinical and biochemical assessments in the post-operative period in assessing the chances of hormonal recovery.
5.8 Strengths and limitations

The strength of this study is that I have sought to clarify specific questions relating to ISP and its relationship to adenoma size and hormonal outcomes in a selected population with NFPMAs, whom are the group at greatest risk of hormone deficiencies. This is an important undertaking given the limited literature available for this group of patients.

The major limitation to this study was the low number of pre-menopausal females recruited. The peak incidence of NFPMAs occurring within late middle age means that few cases occur in pre-menopausal females. Due to the small number of cases in this study, with only five pre-menopausal females (of whom three had ISP data available, and one of which had apoplexy), is was impossible to show statistically significant relationships between the parameters measured. Nonetheless, given this study is exploratory in nature one it still suggests that ISP has possibly some relationship to pre-operative hormone deficiencies. To confirm this finding, this study needs to recruit more participants particularly pre-menopausal females which is achievable through a longer recruitment period beyond this thesis.

Secondly, ISP was unable to be recorded in 15% of cases due to either equipment malfunction or adenoma extravasation. Cystic and haemorrhagic adenomas were likely to extravasate so the pressure in these adenomas could not always be measured. The above issue is likely to be a limiting factor for suitable case recruitment even in larger studies.
5.9 Conclusions

This study set out to investigate ISP and its relationship to adenoma size and ultimately what role it may play in contributing to hormone deficiencies in NFPMAs; relationships that are not well described to date. The findings of this study have shown that ISP does not appear related to adenoma size in NFPMAs but it may possibly have a role in contributing to hormone deficiencies in patients at the time of presentation. These findings do appear so far consistent with what is described in the limited literature in mixed types of pituitary adenomas.

This study did not find any relationship between ISP and adenoma size in NFPMAs however it could not exclude that ISP may be lower in smaller NFPMAs as very few of these occurred in the cohort due to the insufficient recruitment of pre-menopausal females whom are known to have smaller adenomas. Also, ISP is unknown in non-functioning microadenomas as these do not require surgical intervention. In order to confirm these findings a larger sample size is needed as this will give a more accurate representation of the range of adenoma sizes that present for surgery. It may be that there is no relationship between ISP and adenoma size in NFPMAs especially given ISP has also been found to be raised in functioning microadenomas and macroadenomas from this series and from the literature.

This study does suggest however that higher ISP may be a factor associated with hormone deficiencies at presentation in NFPMAs; this finding supported by a very small body of evidence in the literature. This finding requires confirmation by studying a larger cohort with NFPMAs. Adenoma size is also an important factor in hormone deficiencies in NFPMAs. What remains to be defined is the interaction of ISP and adenoma size leading to hormone deficiencies. Quantifying these factors may turn out to be of clinical usefulness in managing cases of NFPMAs. If both ISP and adenoma size are major factors in leading to hormone deficiencies then this strengthens the argument for the consideration of operating in cases of NFPMAs earlier in order to maintain and preserve hormone function rather than a “watch and wait” approach.
Lastly, the role of ISP in apoplexy warrants further exploration given the high prevalence of hormone deficiencies and that ISP has been found to be extremely elevated in a small case series. It may turn out that the clinical usefulness of measuring ISP in apoplexy cases in routine practice may be of greatest benefit if it is able to predict with high likelihood the chances of hormonal losses and recovery. This would require longer recruitment and via a multicentre approach given apoplexy is a rare event.

Presented here is the first data of ISP to be described in a select cohort of NFPMAs: this is of relevance due to the high prevalence of hormonal deficiencies which occur in this type of pituitary adenoma. In order to confirm these findings ongoing recruitment is required. If confirmed, then further analyses can be undertaken to understand the contribution ISP may have in leading to hormone deficiencies in NFPMAs. Ultimately such knowledge may translate to improved hormonal outcomes for those with NFPMAs.
Chapter 6: Summary of the thesis findings and their significance

6.1 Introduction

The research conducted in this thesis has aimed to address several clinical issues relating to the multidisciplinary care of patients with prolactinomas and non-functioning pituitary macroadenomas (NFPMAs) that are central to the multidisciplinary care of these patients. Work from this thesis provides local Australian data from a dedicated pituitary centre (St Vincent’s Hospital Melbourne, Australia) that are of value to patients and doctors from the hospital and wider community and contributes to the international study of pituitary disorders. These attributes fulfil the aims of “pituitary centres of excellence” by providing comprehensive care and support to patients, providing doctor training, continuing medical education and contributing to research in pituitary disorders (171) (179).

6.2 Key findings: Cabergoline-associated valvulopathy in prolactinoma patients

Chapter 2 consist of a collection of works that have evolved over time concerning the association of cabergoline therapy and valvular heart disease in prolactinoma patients; a concern that was first raised over a decade ago (2007) and currently remains an issue due to limited data and lack of consensus about optimal screening methods. Throughout the last decade multiple small studies of prolactinoma cases have been published and have reported on non-specific changes to the cardiac values on echocardiograms, apart from one study that did specifically distinguish the cardinal features of cabergoline-associated valvulopathy (CAV) (49). At the time of starting this thesis the risk of in prolactinoma patients was unknown and screening recommendations for CAV were limited. My thesis works towards addressing this gap.
My initial work consisted of a clinical study and systematic review of the literature leading to recommendations on screening for cabergoline-associated valvulopathy (CAV) (Publication 1). In a local cohort I examined the prevalence of CAV by echocardiograms but also assessed the utility of using a cardiovascular examination as a screening tool; no study to date had reported on the clinical findings of a cardiovascular examination in their cohorts. There were no confirmed cases of CAV in this first Australian cohort. In the systematic review of the literature, of which over 20 different papers had been published, I was able to confirm that these studies were reporting on valvular changes that were not compatible with CAV as these studies had not appreciated the specific echocardiographic findings defining CAV. By highlighting the features of CAV I was able to identify true cases of CAV and found two cases out of 1811 patients (0.11%); the first prevalence data published of this condition.

At that time of this thesis the United States FDA and product disclosure statement recommended echocardiographic screening of individuals prescribed cabergoline at least once or twice per year. The conclusions from my examination of the literature was that this frequency of echocardiograms was not evidence based and unnecessary. I also highlighted limitations to echocardiograms as a screening tool. I made recommendations based on available evidence that annual cardiovascular examination was very effective at excluding a clinically relevant murmur and that echocardiogram should be reserved for those patients with an audible murmur, those treated for more than five years at a dose of more than 3mg per week or those who maintain cabergoline treatment after the age of 50 years. However, the ongoing optimal frequency of echocardiograms was not able to be determined. By following this recommendation of an annual cardiovascular examination the third case of CAV was discovered at the local centre (Publication 2). This is the case of a 52 year old female patient with prolactinoma who required prolonged treatment with high dose cabergoline. The cumulative dose of cabergoline being comparable to the median cumulative dose associated with valvular heart disease in Parkinson’s disease cases reported in the literature. On screening cardiovascular examination, as recommended by my publication (Publication 1), a murmur had been detected leading to a diagnostic
echocardiogram that confirmed the cardinal features of CAV. This case publication, therefore, reinforces my recommendation from Publication 1.

At the time of completion of this thesis in 2019 two recent publications became available from the same British author group demonstrating that even after a decade from the concern of CAV some controversies still exist regarding diagnosing and screening for this condition. The first publication is a meta-analysis of studies of valvular heart disease in prolactinoma cases and the second publication being the first formal positional statement and recommendations for screening for CAV (54) (45). Based on the findings of my systematic review in this thesis I have challenged some of the conclusions of the meta-analysis and published my opinion as a “Letter to the Editor” (Publication 3). In this letter I questioned the authors’ conclusion that cabergoline appeared to be associated with increased prevalence of tricuspid regurgitation by reiterating that non-specific changes in the cardiac valves were being reported on rather than the salient echocardiographic features of CAV. Furthermore, none of the three confirmed cases of CAV in prolactinoma cases have involved disease of the tricuspid valve. I also challenged the authors with their suggestion of “publication of large-scale, prospective, controlled, quality-assured echocardiographic studies with centralized standardized interpretation are needed to provide data and that additional cross-sectional studies drawn from routine practice are unlikely to inform the field significantly”. Given CAV is a rare condition the need for a control group recruited from the healthy population not taking cabergoline is unnecessary for detecting the disease specific condition of CAV as this can only occur in an exposed group. The second publication, the joint positional statement, makes several pertinent comments, however some of these are discordant with comments from the meta-analysis which is surprising given these two publications share some of the same authors. The joint position statement highlights the limitations of echocardiograms, the importance of distinguishing the cardinal features of CAV and that “ongoing collection of high-quality data, via collaborative audit and study initiatives, together with post-marketing reporting,....of independently confirmed cases, is strongly encouraged” (45). These are comments I do agree with. I do however question the validity of their recommendations regarding the frequency of echocardiograms, that clinical cardiovascular examination is not advocated due
insufficient evidence of its usefulness and that in cases of CAV consideration can be made to change treatment to bromocriptine (another ergot dopamine agonist).

6.3 Key findings: Surgical and hormonal outcomes in operated cases of non-functioning pituitary macroadenomas at a single Australian centre

Chapters 3 and 4 of this thesis have examined a large retrospective series of surgically operated cases of NFPMAs at a single centre at St Vincent’s Hospital, Melbourne, Australia, focusing on surgical and hormonal outcomes. Extending on from this Chapter 5 prospectively explores the relationship of ISP to adenoma size and hormonal outcomes in a small cohort of cases with NFPMAs.

Chapter 3 describes surgical outcomes of NFPMAs from a specialist centre. The first finding of interest is that even in expert hands residual disease is commonly encountered post-operatively (61%); a finding similar to other single centre series reported in the literature. Regrowth was observed in 40% of those with residual disease but in those without residual disease recurrence occurred in only 12.5% after a median follow-up of 4 years. Larger baseline adenoma size was a predictor of regrowth and recurrence ($p=0.05$) however that independent association between adenoma size as a predictor of regrowth and recurrences could not be tested in a multivariate analysis due to 19 cases with missing data points. In a multivariate analysis it was found that risk factors for adenoma regrowth and recurrence included the presence of residual disease and younger age at presentation (especially if aged under 41 years).

There are several implications from these findings. Firstly, understanding the surgical outcomes at one’s own centre is important for providing optimal care and counselling for patients at the treating centre. Secondly, to ensure surgical outcomes are comparable to the best international series. Thirdly, appreciating that smaller adenoma size at presentation is
advantageous to better surgical outcomes is an important clinical consideration when planning the optimal time for surgery. Lastly, guidelines on NFPMAs have not been able to define how long to radiologically screen for regrowth and recurrence. Given my finding of an age cut off below 41 years being a risk factor for regrowth and recurrence I have recommended that indefinite radiological screening be considered in younger cases.

Chapter 4 describes hormonal outcomes of NFPMAs from a specialist centre. The novel finding of differences in the frequency of hormone deficiencies between genders has not been previously reported. At presentation it was found that males were more likely to have two or more hormone deficiencies (MHD) as compared to females and post-operatively, very few pre-menopausal females had MHD. The second novel finding was that pre-menopausal females were more likely to recover their reproductive and ACTH-cortisol status which is not seen in males (nor post-menopausal females). In a multivariate analysis that included factors associated with post-operative hormone deficiencies adenoma size was found to be a significant factor. In this study pre-menopausal females did have smaller adenomas compared to males and males of the same age, thus explaining why they had fewer hormone deficiencies.

There are several implications from these findings. Firstly, (and similar to the surgical outcomes study from Chapter 3), understanding the hormonal outcomes of patients at one’s own centre is important for providing optimal care and counselling for patients at the treating centre. Secondly, there should be little reservation in offering pituitary surgery to pre-menopausal women as they have favourable hormonal outcomes which is of clinical importance to their reproductive status. Thirdly, appreciating that smaller adenoma size at presentation is a factor in optimal hormonal outcomes is an important clinical consideration when planning the optimal time for surgery.

In finding that pre-menopausal females with NFPMAs had such favourable hormonal outcomes post-operatively I wanted to explore how intrasellar pressure (ISP), another less
well described factor, relates to hormone deficiencies in NFPMA
s as there is a paucity of
information about this. In Chapter 5 of my thesis I set out to understand ISP and explore its
relationship to adenoma size and hormonal outcomes in NFPMAs. In a cohort of 41 cases a
relationship between ISP and adenoma size in NFPMAs was not found. However, it could
not exclude that ISP may still be lower in smaller NFPMAs as very few of these occurred in
the cohort due to the low number of pre-menopausal females, who are known to have
smaller adenomas. This study does however suggest that higher ISP may be associated with
hormone deficiencies at presentation in NFPMAs: the p value of 0.088 might indicate a Type
2 statistical error with insufficient numbers to show a significant difference. This would be
consistent with two studies in the literature that have shown higher ISP in those with
hormone deficiencies at presentation (113) (185).

Adenoma size again emerged as a potentially important factor in hormone deficiencies in
NFPMAs in this cohort but again this failed to reach statistical significance (p=0.079). What
currently remains unexplored is the complex interaction of ISP and adenoma size leading to
hormone deficiencies. By confirming the above findings in a larger study further statistical
methods could be used to investigate how these interactions can result in hormone
deficiencies. Ultimately by understanding the pathophysiology of hormonal deficiencies in
NFPMAs in greater detail than what is currently understood may translate to methods to
improve outcomes for these patients.

The role of ISP in apoplexy is an area of interest but due to the rarity of this event only two
cases of apoplexy occurred in this cohort. Apoplexy is known to be associated with a high
prevalence of hormone deficiencies due to pituitary tissue destruction; apoplexy has been
associated with extreme elevation ISP in a small case series. It may turn out that the clinical
utility of measuring ISP in apoplexy cases in routine practice may be of greatest benefit if it
is able to assist in predicting the chances of hormonal loss and recovery. This would require
a study with a longer recruitment period and via a multicentre approach given apoplexy is a
rare event.
From these studies of NFPMAs in this thesis adenoma size emerges as an important factor that influences surgical and hormonal outcomes. In addition I have observed that there are several differences in the cohorts of NFPMAs studied in this thesis that further supports the influence of adenoma size. The recently studied cohort from 2015-2018 (Chapter 5) were more likely to have presented incidentally, had significantly smaller and less invasive adenoma and were more likely to be eutuitary post-operatively compared to the historical cohort of 1995-2010 (Chapter 4). The differences observed between these cohorts are highly suggestive that in more recent times cases of NFPMAs may be presenting earlier in the natural history of the disease, in turn, resulting in favourable patient outcomes. Authors Messerer et al., have shown previously in a series of 76 cases of NFPMAs that those who presented truly asymptomatically (no evidence of mass effect or hormone deficiencies) had significantly smaller adenomas, lower prevalence of residual disease and excellent hormonal outcomes post-surgery compared to cases that presented symptomatically (204). The authors from this study above concluded that it is appropriate for physicians and experienced neurosurgeons to consider surgical treatment in asymptomatic cases as the outcomes are better (204). It is well recognised that remission rates in surgically treated functioning adenomas are highest when adenomas are smaller at the time of surgery (121) (122) (134) but in the case of NFPMAs the current and established indication for surgery is when these adenomas are causing mass effect or when hormone deficiencies occur (56). The overall findings of my thesis would support an early consideration of surgery in cases with NFPMAs rather than a “watch and wait” approach in order to optimise both surgical and hormonal outcomes for these patients.
7.0 Conclusion and future directions

In this thesis I have explored several important aspects of the two most prevalent adenomas types of prolactinomas and NFPMAs made possible by studies undertaken at a specialist pituitary centre, St Vincent’s Hospital, Melbourne, Australia. Some findings have been novel and other findings have confirmed what has been previously described but all these findings are of clinical utility in the daily management of patients with these pituitary adenoma types. These data have an impact from counselling patients at the onset of their condition, throughout their long-term management and are relevant to endocrinologists and neurosurgeons who are involved in multidisciplinary care of patients with these adenomas. The data presented are relevant to the local practice, are also the first major Australian contribution in the specific subject areas presented in this thesis as well as contributing to the international literature. These contributions also help to fulfil core aims of being a pituitary “centre of excellence”. Additionally, this thesis highlights changes in time with evolving literature in the subject of CAV and to the demographic presentation of cohorts of NFPMAs. These changes demonstrate the need for ongoing research in centres that focus on the care of pituitary patients.

My findings from examining the issue of CAV in prolactinoma patients came at a time when there the literature was reporting non-specific findings on echocardiograms and recommendations for screening had no evidence base. My work clarified the salient features of CAV on echocardiogram, reported on the prevalence of CAV and made recommendations on screening for CAV.

Over a decade has passed since the issue of CAV was first raised, yet it is evident from following the literature that data are still be being acquired and that optimal screening recommendations and management are still evolving. As CAV is a rare event in prolactinoma patients it is likely that another decade will pass before further information becomes available that will significantly change the field. My thesis highlights several areas that still
need to be explored going forward. Firstly, that it is important for each multidisciplinary team to decide on suitable screening strategies for their own patients especially when guidelines for screening for CAV are limited and are controversial. Secondly, it is vital that centres maintain and publish ongoing data of their cohorts as this is the best way of finding the true prevalence of CAV. Thirdly, aiming to reduce a patient’s lifetime exposure to cabergoline, by using the minimum effective dose and for the shortest period of time, is an important message that is yet to be emphasised and needs to be full explored. Lastly, novel treatments for prolactinomas need to be investigated as alternate treatment options to cabergoline.

Several novel findings have emerged from my studies examining surgical and hormonal outcomes of operated cases of NFPMAs which impact on the management of the NFPMAs from a local and international perspective. These include the risks of regrowth and recurrence associated with younger age, gender differences in hormonal outcomes and the particularly favourable hormonal outcomes seen in pre-menopausal females. Collectively, these studies have all noted that smaller adenoma size is a key factor in favourable surgical and hormonal outcomes. Furthermore, the observation that the later cohort of NFPMA studied in this thesis had smaller adenomas than the historical cohort strongly suggests that cases of NFPMAs may be now presenting earlier in the disease process which appears to be advantageous.

The significance of smaller adenomas being associated with favourable outcomes highlights several areas that still need to be explored going forward. Firstly, to strive to educate the endocrine and neurosurgical community of the value of considering surgical intervention in cases of NFPMAs sooner rather than later can improve surgical outcomes and preserve hormone function. It is well recognised that remission rates for surgically treated functioning adenomas are optimised when smaller adenomas are treated however this is not the case with NFPMAs whereby the primary indication for surgery is when lesions are causing mass effects or hormone deficiencies have occurred. Secondly, to explore methods by which NFPMAs can be diagnosed earlier in the disease process by raising awareness of
pituitary disorders in the community and education of primary care physicians. Thirdly, that further molecular and genetic discoveries about the pathogenesis of NFPMAs may evolve to be the key for understanding the variable clinicopathological behaviour of NFPMAs which will, no doubt, change the way that these adenomas are managed. Lastly, it is paramount that ongoing review of outcomes of large cohorts with NFPMAs continue in order to detect clinically important changes over time; this information that will undoubtedly result in improved patient outcomes. Such reviews are possible though “pituitary centres of excellence” where large cohorts can be studied in an appropriate fashion.

Finally, raised ISP may possibly be an important factor in the development of hormone deficiencies although this point in time my thesis has been unable to quantify the relationships between ISP, adenoma size and hormone deficiencies. In order to clarify these relationships recruitment of a larger cohort is required; this approach is being continued beyond this thesis.
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Appendices

Appendix 1: Results of ISP in functioning pituitary adenomas

Demographic results in functioning pituitary adenomas

In total 23 cases were recruited for this study of which five cases were excluded: two cases being other types of pituitary tumours (a spindle cell oncocytoma and a meningioma) and a further three cases were excluded due to ISP equipment issues (one case due to a faulty electrical cable for the monitor, one case where the ISP probe failed to take a measurement and one case where the ISP probe could not be located in operating room). Data was therefore available on 18 cases of functioning pituitary adenoma. There were no cases of apoplexy in the functioning adenoma cohort.

Cases consisted of seven males and 11 females (seven post-menopausal females and four pre-menopausal females) with mean age of 46.1 years (13.4) (range 21-71). Adenoma type consisted of somatotropinomas (10), corticotropinomas (6), one prolactinoma and one TSHoma. Macroadenomas occurred in 11 cases, microadenomas in five cases and two cases had non-lesional adenomas (both were corticotropinomas which were confirmed by histological staining for ACTH) (Table 1.0).

Adenoma size (both by maximal diameter and volume) in those with functioning adenomas tended to be larger in males compared to females (p=0.07) (Table 1.0).

ISP results in functioning pituitary adenomas

In 18 cases of functioning adenoma, median ISP was 22.5mmHg (14.5, 32) (range 8-57). ISP was compared between somatotropinomas and corticotropinomas with no difference
between median ISP detected (Table 1.1). The highest ISP readings of 57mmHg and 52mmHg were found in a pre-menopausal female with a corticotropic microadenoma and a post-menopausal female with a somatotropic macroadenoma, respectively.

Median ISP in patients with microadenomas and macroadenomas were not significantly different (19mmHg vs 23mmHg, respectively). In two cases of non-lesional adenomas ISP were 12mmHg and 28mmHg (Table 1.1). Figure 1.0 shows the plot of ISP by categorical size in functioning adenomas. There was no correlation found between ISP and maximal adenoma diameter or volume in functioning adenomas (Table 1.1). In addition, there was also no correlation found between ISP and volume in functioning adenomas and NFPMAs combined (Table 1.1).

Figure 1.1 shows the plot of ISP by adenoma categorical size of NFPMAs combined with functioning adenomas with no significant differences being detected between categorical sizes.

Only 2/18 cases of functioning adenomas were invasive. One case was in a male with prolactinoma exhibiting cavernous sinus invasion and an elevated ISP of 38mmHg. The other case was also a male with a TSHoma with a nubbin of adenoma projecting into the arachnoid space in the brain; the ISP reading in this case was 23mmHg.

There was no difference in ISP between males and females (23mmHg vs 20mmHg, respectively, p=0.5). Median ISP was 25.5mmHg in pre-menopausal females; a value that was not different to males and post-menopausal females (Table 1.1).

With increasing age ISP appeared to be lower in those with functioning adenomas, although was not significant, p=0.095 (Table 1.1). This relationship of ISP and age was not found in those with NFPMAs.
### Table 1.0 Demographic data of functioning adenomas

<table>
<thead>
<tr>
<th>Gender Subgroup</th>
<th>All</th>
<th>Males</th>
<th>Females</th>
<th>Pre-MenF</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>18</td>
<td>7</td>
<td>11</td>
<td>4</td>
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<tr>
<td><strong>Age at presentation</strong>&lt;br&gt;(years); mean (SD)&lt;br&gt;(range 21-71)</td>
<td>46.1 (13.4)</td>
<td>41.4 (5.9)</td>
<td>49.1 (16.3)</td>
<td>32.2(12.4)</td>
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<tr>
<td>Adenoma type</td>
<td></td>
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</tr>
<tr>
<td>Somatotropinoma</td>
<td>10</td>
<td>5</td>
<td>5</td>
<td>1</td>
</tr>
<tr>
<td>Corticotropinoma</td>
<td>6</td>
<td>1</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Prolactinoma</td>
<td>1</td>
<td>1</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>TSHoma</td>
<td>1</td>
<td>1</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td><strong>Adenoma size</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Non-lesional</td>
<td>2</td>
<td>2</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Microadenoma</td>
<td>5</td>
<td>1</td>
<td>5</td>
<td>0</td>
</tr>
<tr>
<td>Macroadenoma</td>
<td>11</td>
<td>11</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td><strong>Volume (mm³)</strong></td>
<td>492 (129, 1948)</td>
<td>919 (419, 7959)</td>
<td>190 (0, 1885)</td>
<td>217 (73, 1270)</td>
</tr>
<tr>
<td><strong>Maximum size (mm): median (IQR)</strong></td>
<td>11 (7.1, 17)</td>
<td>15 (10, 27)</td>
<td>8.9 (4.8, 16)</td>
<td>8.9 (5.5, 15.3)</td>
</tr>
<tr>
<td>Invasive adenoma</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cavernous sinus</td>
<td>1</td>
<td>1</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Infraesellar</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Intracranial</td>
<td>1</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td><strong>Hormonal Status</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Presentation</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>0 HD</td>
<td>14 (77.8%)</td>
<td>3 (16.6%)</td>
<td>1 (5.6%)</td>
<td></td>
</tr>
<tr>
<td>1 HD</td>
<td>1 (5.6%)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>MHD (≥2HD)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Pre-menopausal females; Pre-MenF, HD: Hormone deficiency, MHD: Multiple hormone deficiency
Table 1.1 ISP in functioning adenomas

<table>
<thead>
<tr>
<th></th>
<th>Intrasellar Pressure (mmHg);</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>median (IQR)</td>
<td></td>
</tr>
<tr>
<td>All</td>
<td>22.5 (14.5, 32)</td>
<td>Range 8-57</td>
</tr>
<tr>
<td><strong>Gender</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Males n=7</td>
<td>23 (20-31)</td>
<td></td>
</tr>
<tr>
<td>Females n=11</td>
<td>20 (12-35)</td>
<td>p=0.62</td>
</tr>
<tr>
<td>Pre-menopausal females n=4</td>
<td>25.5 (14.8, 50.5)</td>
<td>† p=0.63</td>
</tr>
<tr>
<td><strong>Age (years)</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>21-41</td>
<td>27 (14.5, 37.5)</td>
<td></td>
</tr>
<tr>
<td>42-50</td>
<td>24 (20.0, 30.5)</td>
<td></td>
</tr>
<tr>
<td>51-71</td>
<td>15.5 (8.75, 39.7)</td>
<td>p=0.48</td>
</tr>
<tr>
<td>Continuous variable</td>
<td></td>
<td></td>
</tr>
<tr>
<td>(correlation co-efficient with 95% CI)</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>-0.34 (-0.85, 0.17)</td>
<td>p=0.18</td>
</tr>
<tr>
<td><strong>Adenoma Type</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Somatotropinoma</td>
<td>21 (13.5, 32)</td>
<td></td>
</tr>
<tr>
<td>Corticotropinoma</td>
<td>24 (12.8, 37.5)</td>
<td>p=0.8</td>
</tr>
<tr>
<td><strong>Adenoma size</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Non-lesional (n=2)</td>
<td>20</td>
<td></td>
</tr>
<tr>
<td>Microadenoma (n=5)</td>
<td>19 (10.5, 44)</td>
<td></td>
</tr>
<tr>
<td>Macroadenoma (n=11)</td>
<td>23 (20, 35)</td>
<td>p=0.9</td>
</tr>
<tr>
<td>Maximum diameter</td>
<td></td>
<td></td>
</tr>
<tr>
<td>(correlation co-efficient with 95% CI)</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>0.26 (-0.29, 0.81)</td>
<td>p=0.34</td>
</tr>
<tr>
<td>Volume</td>
<td></td>
<td></td>
</tr>
<tr>
<td>(correlation co-efficient with 95% CI)</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>0.08 (-0.09, 0.26)</td>
<td>p=0.35</td>
</tr>
<tr>
<td>Volume (All adenomas included)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>(correlation co-efficient with 95% CI)</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>0.05 (-0.06, 0.16)</td>
<td>p=0.38</td>
</tr>
</tbody>
</table>

† Kruskal-Wallis for gender subgroup difference p=0.63
† All adenomas included: functioning adenomas and NFPMAs
Figure 1.0 Plot of ISP in functioning adenomas by categorical size

Data plot of ISP in functioning adenoma by categorical size of non-lesional, microadenomas and macroadenomas. Horizontal line representing median ISP in each group. In two cases of non-lesional adenomas ISP was 12mmHg and 28mmHg (median 20 mmHg), in microadenomas median ISP was 19mmHg and macroadenomas ISP was 23mmHg. No significant differences in median ISP were found between groups (p=0.9).
Figure 1.1 Plot of ISP in all pituitary adenomas combined (NFPMAs and functioning adenomas) by categorical size

Data plot of ISP in all pituitary adenomas combined (NFPMAs and functioning adenomas) by categorical size. Horizontal line representing median ISP in each group. In two cases of non-lesional adenomas ISP was 12mmHg and 28mmHg (median 20mmHg), in microadenomas median ISP was 19mmHg and in macroadenomas ISP was 22mmHg. There were no significant differences in the median ISP between groups (p=0.9).
Appendix 2: Results of other analyses in NFPMAs

Hormonal outcomes

Of the cohort of 35 with ISP data, 11 (31.4%) of cases had no hormone deficiency at presentation and only 2/11 (18%) developed a new hormone deficiency. Of the remaining 24 (68.6%) of cases who had one or more hormone deficiencies at presentation, 11/24 (45.8%) recovered a hormone axis and 13/24 (54.2%) did not recover any hormone axis. In this later group who did not recover any hormonal axis there were two cases that lost an additional hormonal axis. In total there were four cases that developed a new hormonal axis deficiency.

ISP in individual hormonal axis deficiencies

The relationship between ISP and individual hormonal axis losses at presentation and post-operatively was also examined. The prevalence of each axis deficiency and median ISP found in groups with and without the individual hormone axis deficiency are shown in Table 2.0 A and B, respectively at presentation, and shown in Table 2.1 A and B, respectively for the post-operative period. No statistically significant differences in median ISP were found in those cases with the individual hormonal axis compared to those without the particular axis deficiency.

Pre-operative and post-operative hormonal status and adenoma volume

Median adenoma volume in those with no hormone deficiency pre-operatively tended to be smaller than those with one hormone deficiency and MHD (2420mm$^3$ vs 3935mm$^3$ and 3044mm$^3$, respectively) although trend analysis did not demonstrate statistical significance (p=0.16). Median adenoma volume in those with no hormone deficiency pre-operatively
tended to be smaller than those with any hormone deficiency although this did not reach statistical significance (2420mm$^3$ vs 3551mm$^3$, p=0.079) as shown in Table 2.2.

Median adenoma volume in those with no hormone deficiency post-operatively appeared to be smaller than those with any hormone deficiency and MHD (2693mm$^3$ vs 3820mm$^3$ and 4343mm$^3$, respectively) but trend analysis did not demonstrate statistical significance (p=0.15). Median adenoma volume in those with no hormone deficiency post-operatively tended to be smaller than those with any hormone deficiency although this did not reach statistical significance (2693mm$^3$ vs 3820mm$^3$, p=0.081) as shown in Table 2.2.

**Hormone recovery and losses**

Median adenoma volume was 2809mm$^3$ in those that recovered an axis and 4299mm$^3$ in those that did not recover but this difference was not statistically significant (p=0.3) (Table 2.3).

In those that developed a new hormone deficiency adenoma volume tended to be larger than those whom did not develop a new hormone deficiency (5620mm$^3$ vs 2693mm$^3$ respectively, p=0.066) as shown in Table 2.3.

Prolactin levels in those that recovered a hormonal axis was not different from those that did not recover the respective axis (Table 2.3).
**Table 2.0 A and B. Pre-operative hormone deficiency and ISP in 35 cases NFPMA**s

A: Frequency of pre-operative hormone deficiencies in 35 cases with ISP data

<table>
<thead>
<tr>
<th>Hormonal axis</th>
<th>All 35</th>
<th>Male 20</th>
<th>Female 15</th>
<th>Pre-MenF 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gonadal</td>
<td>24 (68.6%)</td>
<td>16 (80%)</td>
<td>8 (53.3%)</td>
<td>2 (66.7%)</td>
</tr>
<tr>
<td>Thyroid</td>
<td>9 (25.7%)</td>
<td>6 (30%)</td>
<td>3 (20%)</td>
<td>0</td>
</tr>
<tr>
<td>ACTH-cortisol</td>
<td>9 (25.7%)</td>
<td>6 (30%)</td>
<td>3 (20%)</td>
<td>1 (33.3%)</td>
</tr>
</tbody>
</table>

B: Pre-operative hormone deficiency by axis and ISP in 35 cases with ISP data

<table>
<thead>
<tr>
<th>Hormonal axis</th>
<th>ISP (mmHg): median (IQR)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gonadal deficiency vs no deficiency</td>
<td>22 (16, 34) vs 20.5 (11, 24.8) p=0.2</td>
</tr>
<tr>
<td>Thyroid deficiency vs no deficiency</td>
<td>28.5 (19, 37.8) vs 20 (15, 25) p=0.2</td>
</tr>
<tr>
<td>ACTH-Cortisol deficiency vs no deficiency</td>
<td>22 (17, 46) vs 21.5 (14.8, 27.3) p=0.4</td>
</tr>
</tbody>
</table>

Table 2.1 A and B. Post-operatively hormone deficiency and ISP in 35 cases NFPMA**s

A: Frequency of post-operative hormone deficiencies in 35 cases with ISP data

<table>
<thead>
<tr>
<th>Hormonal axis</th>
<th>All 35</th>
<th>Male 20</th>
<th>Female 15</th>
<th>Pre-MenF 3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gonadal</td>
<td>20 (57.1%)</td>
<td>13 (65%)</td>
<td>7 (46.7%)</td>
<td>2 (66.7%)</td>
</tr>
<tr>
<td>Thyroid</td>
<td>11 (31.4%)</td>
<td>8 (40%)</td>
<td>3 (20%)</td>
<td>0</td>
</tr>
<tr>
<td>ACTH-cortisol</td>
<td>4 (11.4%)</td>
<td>4 (20%)</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

B: Post-operatively hormone deficiency by axis and ISP in 35 cases with ISP data

<table>
<thead>
<tr>
<th>Hormonal axis</th>
<th>ISP (mmHg): median (IQR)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gonadal deficiency vs no deficiency</td>
<td>24 (16, 34) vs 19 (14.3, 24.8) p=0.2</td>
</tr>
<tr>
<td>Thyroid deficiency vs no deficiency</td>
<td>27 (13, 34) vs 20.5 (16, 24.5) p=0.6</td>
</tr>
<tr>
<td>ACTH-Cortisol deficiency vs no deficiency</td>
<td>16 (16, 25.8) vs 22 (15, 30) p=0.5</td>
</tr>
<tr>
<td>Recovery of axis vs no recovery</td>
<td>28 (18, 41) vs 22 (16, 28.5) p=0.3</td>
</tr>
</tbody>
</table>
### Table 2.2 Pre- and post-operative hormonal status and adenoma volume in 41 cases of NFPMAs

<table>
<thead>
<tr>
<th>Hormonal status</th>
<th>Adenoma volume (mm³): median (IQR)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Pre-operative n=41</strong></td>
<td></td>
</tr>
<tr>
<td>No HD</td>
<td>2420mm³ (1403, 4461)</td>
</tr>
<tr>
<td>1HD</td>
<td>3935mm³ (2518, 5178)</td>
</tr>
<tr>
<td>MHD</td>
<td>3044mm³ (2171, 7083)</td>
</tr>
<tr>
<td>No HD vs Any HD</td>
<td>2420mm³ (1403, 4461) vs 3551mm³ (2413, 6647)</td>
</tr>
<tr>
<td><strong>Post-operative n=40</strong></td>
<td></td>
</tr>
<tr>
<td>No HD</td>
<td>2680mm³ (2003, 3994)</td>
</tr>
<tr>
<td>1HD</td>
<td>3044mm³ (1846, 10968)</td>
</tr>
<tr>
<td>MHD</td>
<td>4343mm³ (2091, 7351)</td>
</tr>
<tr>
<td>No HD vs Any HD</td>
<td>2693mm³ (674, 4179) vs 3820mm³ (2050, 7351)</td>
</tr>
</tbody>
</table>

HD: Hormone deficiency, MHD: Multiple hormone deficiency

† Cuzick’s test for trend

### Table 2.3 Post-operative hormonal loss and recovery by volume and prolactin level in 41 cases of NFPMAs

<table>
<thead>
<tr>
<th>Hormonal status</th>
<th>Adenoma volume (mm³) median (IQR) or Prolactin (mIU/L) median (IQR)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Recovered HD (n=12) vs no recovery HD (n=13)</td>
<td>2809mm³ (2352, 4439) vs 4299mm³ (2360, 8446)</td>
</tr>
<tr>
<td>Developed new HD vs no new HD</td>
<td>5620mm³ (3597, 11119) vs 2693mm³ (2009, 4402)</td>
</tr>
<tr>
<td>Pre-operative prolactin level if recovered HD vs prolactin level in those not recovered HD</td>
<td>424.5 (260, 748.5) vs 315.5 (183, 496)</td>
</tr>
</tbody>
</table>

HD: Hormone deficiency
Appendix 3: Differences in the cohorts of NFPMAs studied in this thesis

Table 3.0 Differences observed between the historical cohort (Chapter 4) and current cohort (Chapter 5) of NFPMAs studied in this thesis

<table>
<thead>
<tr>
<th>Observation</th>
<th>Historical cohort 1995-2010 n=141</th>
<th>Current cohort 2015-2018 n=41</th>
<th>Confidence interval and p value</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Age (years)</strong> mean (SD)</td>
<td>57.2 (15.8)</td>
<td>60 (12.3)</td>
<td>3.4 (95%CI 1.2, 7.9), p=0.15</td>
</tr>
<tr>
<td><strong>Incidental presentation</strong></td>
<td>31% (43/141)</td>
<td>41.5% (17/41)</td>
<td>p=0.19</td>
</tr>
<tr>
<td><strong>Hormonal status</strong> Presentation</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No HD</td>
<td>28%</td>
<td>34.1%</td>
<td>p=0.44 †</td>
</tr>
<tr>
<td>1 HD</td>
<td>34.4%</td>
<td>29.3%</td>
<td></td>
</tr>
<tr>
<td>MHD</td>
<td>37.6%</td>
<td>36.6%</td>
<td></td>
</tr>
<tr>
<td>Post-operatively</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No HD</td>
<td>29%</td>
<td>47.5%</td>
<td>P=0.047 †</td>
</tr>
<tr>
<td>1 HD</td>
<td>32.3%</td>
<td>22.4%</td>
<td></td>
</tr>
<tr>
<td>MHD</td>
<td>38.7%</td>
<td>30%</td>
<td></td>
</tr>
<tr>
<td><strong>Adenoma size (mm)</strong> median (IQR)</td>
<td>n= 97</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>30 (22, 34) (Range 13-65)</td>
<td>22 (18.5, 27) (Range 13-45)</td>
<td>6 (95% CI 3, 8.4) p=&lt;0.001</td>
</tr>
<tr>
<td><strong>Invasive adenoma</strong></td>
<td>51.3 % (67/132)</td>
<td>26% (10/41)</td>
<td>P=0.004</td>
</tr>
</tbody>
</table>

HD: Hormone deficiency, MHD: multiple hormone deficiency (≥2 hormone deficiency)

† Comparison between cohorts with no hormone deficiencies pre and post-operatively, respectively

‡ Size was compared using maximal diameter as volumetric data was not available in the historical cohort