EXPERT OPINION

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Emerging drugs for Cushing's disease

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Introduction: Considering the effects of uncontrolled hypercortisolism on morbidity and mortality, there is a clear need for effective medical therapy for patients with Cushing's disease (CD). Therefore, the search for new medical effective tools remains active, and already promising results have been obtained.

Areas covered: The importance of the design and conduct of trials to validate old drugs or to test new compounds is discussed. The results of the ongoing clinical trials, targeting the specific properties of drugs, such as ketoconazole, LCI699, mifepristone, etomidate and pasireotide, are also reported. The authors also emphasise the advantages and drawbacks of each particular drug, and the potential combined use of agents with complementary mechanisms of action.

Expert opinion: CD is an excellent example of a situation where effective therapy is essential, but where the balance of risk and benefit must be carefully judged. Metyrapone is the drug of choice when rapid control of the hypercortisolaemia is required, ketoconazole represents a good second-line drug, although in the future LCI699 may be a better alternative. Mifepristone can also be used in the rare situation when previous drugs are inappropriate. Etomidate is useful where immediate parenteral action is required. For drugs working directly on the pituitary, cabergoline is occasionally effective and pasireotide can be attempted in patients with mild CD.

Keywords: Cushing disease, ketoconazole, LCI699, mifepristone, pasireotide

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1. Background

1.1 Definition, pathophysiology and clinical presentation

Cushing's disease (CD), pituitary-dependent Cushing's syndrome, is a rare disorder characterised by the overproduction of adrenocorticotrophin (ACTH) [1]. Excess secretion of ACTH augments cortisol production by the adrenal gland, disturbing the normal cortisol feedback mechanism of the hypothalamo-pituitary-adrenal (HPA) axis, with loss of the circadian rhythm and excess cortisol secretion [2]. After being secreted, glucocorticoids exert their effects through the glucocorticoid receptor (GR), which is expressed in almost every human tissue. Excess glucocorticoids, such as in CD, result in deleterious effects on cell metabolism, such that patients develop the clinical features of CD [3]. The clinical presentation of hypercortisolism varies from patient to patient and includes a wide combination of signs and symptoms, such as weight gain, severe fatigue and muscle weakness, depression, cognitive impairment, purplish skin striae, easy bruising, loss of libido, hirsutism, acne and menstrual disorders. Hypertension occurs in 80% of patients with Cushing's syndrome and is thought to be mostly caused by the combined effect of cortisol on both the GR and the mineralocorticoid receptor, although many other factors may be involved causing enhancement of vasoactive substances and/or a suppression of the vasodilatory systems, or even by effects on cardiovascular regulation through

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the CNS via both glucocorticoid and mineralocorticoid receptors [4]. In some sites, excess cortisol overwhelms the ability of type 2 11β-hydroxysteroid dehydrogenase to convert it to cortisone, and thus cortisol has access to and activates the mineralocorticoid receptor. Impaired glucose tolerance or type 2 diabetes also occurs in 80% of patients as a result of increased insulin resistance and impaired insulin secretion [5]. Glucocorticoids increase liver gluconeogenesis and decrease glucose uptake in skeletal muscle and adipose tissue [5]. Impaired insulin secretion also occurs as a result of glucocorticoids binding to pancreatic β-cells, resulting in impaired β-cell function [5].

While in many patients clinical suspicion of Cushing's syndrome is based on a combination of sensitive (central obesity) and specific (related to protein wasting) signs [6], in other cases the clinical picture is much less clear and confusing. For this reason, efficient screening and confirmatory procedures are essential before considering therapy. Measuring serum cortisol resistance to dexamethasone, or in blood or saliva at midnight, are the most effective ways to detect hypercortisolism. The final diagnosis, and the search for its cause, requires sophisticated hormonal testing and imaging procedures best performed in specialised referral centres [6].

1.2 Approach to the patient with CD

Once the diagnosis of CD is established, treatment should aim to reverse of clinical features, normalise biochemical changes with minimal morbidity, and to lead to an improvement in patient well-being, and long-term control of the disease and the tumour without recurrence [7]. Treatment of all comorbidities also plays an important role [7].

Transsphenoidal surgery (TSS) is the recommended first-line treatment of CD [7]. TSS is potentially curative, resulting in remission of CD in about 65 – 90% of patients with a pituitary microadenoma, is associated with only a small risk of complications if performed by an experienced surgeon and is generally associated with preservation of pituitary function [7]. However, even among those who achieve remission post-operatively, a recurrence rate of 10 – 20% has been reported in patients followed for more than 5 and 10 years [8,9]. Repeat TSS may be required in these patients with persistent or recurrent disease to achieve remission in the event of tumour recurrence, but it is associated with lower remission rates and higher rates of hypopituitarism [10,11]. Other treatments options include pituitary radiotherapy and bilateral adrenalectomy, although these are associated with significant lifelong complications [12].

Radiotherapy (RT) may be recommended for residual or recurrent disease when further TSS is not appropriate. RT is also performed to prevent or treat Nelson's syndrome (NS), corticotroph tumour progression. The rate of remission of CD after RT reported in the literature varies considerably [7]. In a recent study, disease remission at 1, 2, 3 and 5 years after 'gamma knife' therapy in 96 patients with CD was 34, 54, 72 and 78%, respectively [13]. RT has also been shown to be effective in controlling tumour growth in more than 90% of patients [14].

Bilateral adrenalectomy is still used as a definitive treatment for CD, especially in patients with severe CD, as it will control hypercortisolaemia immediately. Another important indication for bilateral adrenalectomy is the treatment of CD resistant to surgery in women in the reproductive age, seeking fertility without stimulation of ovulation, as gonadotrophin deficiency may occur after RT. Bilateral adrenalectomy was traditionally performed through an open approach, but since the advent of laparoscopic adrenalectomy the laparoscopic approach has become the surgical method of choice. Advances in technology and refinement in surgical skills have greatly reduced morbidity and mortality associated with adrenalectomy surgery in such a high-risk patient population. However, this treatment requires life-long replacement of glucocorticoids and mineralocorticoids and carries an increased standardised mortality ratio of about 2.0 [15]. Moreover, adrenalectomy does not treat the pituitary adenoma itself, and because of the risk of developing NS, MRI scans and ACTH evaluation have to be performed regularly [16]. Occasionally, adrenal tissue may regrow under the stimulatory action of chronically elevated ACTH plasma levels [6].

Contrary to other pituitary adenomas such as prolactinomas [17] or growth hormone (GH)-secreting adenomas [18] in which medical treatment has become important, the medical treatment of CD has traditionally had a marginal role. Nevertheless, there are numerous circumstances in which the medical treatment of CD may be indicated.

2. Medical need

There are several indications for medical therapy in CD (**Table 1**). The rationale for medical treatment may vary considerably, ranging from a temporary palliative measure to employing the last available resources and may be an intermediary measure in the management of difficult cases.

2.1 Mobility and mortality of CD

The high morbidity and mortality caused by hypercortisolism justifies the need for effective drugs to control the disease. Untreated ACTH-dependent Cushing's syndrome is associated with a risk of life-threatening cardiovascular, infectious and metabolic complications [2,19,20] and is associated with severe morbidity. Chronic hypercortisolism induces major changes in body composition with abdominal and facial fat accumulation, muscle and skin atrophy, and osteoporosis [21]. Excess cortisol has also major effects on the brain that can result in psychopathology and neurocognitive dysfunction [22]. Approximately 55 – 80% of CD patients have major depression or an anxiety disorder [23]. Moreover, as mentioned earlier, CD is accompanied by all components of the metabolic syndrome, including overweight/obesity, hypertension, impaired glucose tolerance/diabetes and dyslipidaemia [21]. CD is associated with increased mortality, largely due to the high

Table 1. Indications for medical therapy in Cushing's disease.

During preparation for surgery
Patients with contraindication for surgery or a high operation risk
Patients unwilling to undergo surgery
Patients with low probability of surgical cure (unfavourable localisation, invisible adenomas…)
Control of metabolic effects of hypercortisolemia in patients with potential life threatening complications
After unsuccessful surgery or recurrence of the disease
Patients waiting for radiation therapy to take effect
Whenever a definitive treatment is delayed
Metastatic disease

cardiovascular risk. Hypercortisolism induces vascular damage both directly and indirectly, favouring the occurrence of premature atherosclerosis [24]. Furthermore, apart from an increased risk of arterial thrombosis, CD is also associated with an increased risk of venous thromboembolic disease due to both activation of the coagulation cascade and impaired fibrinolysis [25]. Finally, miscellaneous features of CD include hirsutism, gonadal dysfunction, nephrolithiasis and (most importantly) increased susceptibility to infection [1].

Depending on their action site, the various medical drugs available for CD can be classified into three groups: agents that inhibit adrenal steroidogenesis; drugs that directly target ACTH secretion and ligands that block the cortisol receptor (Table 2) [26-28]. Although many drugs have been used 'off-label' to treat CD, currently there are four approved therapeutics for CD. Pasireotide (*Signifor*, Novartis) is the only medical therapy approved in the European Union and the US and is licensed for use in patients for whom surgery is not an option or has failed [29,30]. Mifepristone (*Korlym*), a GR antagonist, is approved in the US for the control of hyperglycaemia secondary to hypercortisolism, in patients with endogenous Cushing's syndrome who have type 2 diabetes mellitus or glucose intolerance and have failed or are not candidates for surgery [31]. Recently, at least in Europe, there is an on-going expansion of the licences for metyrapone (*Metopirone*) and ketoconazole (*Ketoconazole HRA*). Hopefully, this number is likely to rise with the initiation of new studies and completion of on-going clinical trials.

3. Existing treatment

3.1 Adrenal steroidogenesis inhibitors

Since CD is caused by a pituitary tumour, medical therapy should ideally target the corticotroph cell adenoma. However, as glucocorticoids represent the final product of the hypothalamic-pituitary-adrenal axis, steroidogenic inhibition was the first therapy to be used. Adrenal enzyme inhibitors inhibit one or more enzymes involved in cortisol synthesis. Combinations of these drugs often have additive or synergistic therapeutic effects at lower individual doses, thereby minimising adverse effects. However, the availability of these drugs varies in different countries. In the US, the most commonly used drug is ketoconazole; in the UK, metyrapone has been used more commonly. Both metyrapone and ketoconazole, marketed by HRA Pharma, have recently been approved for the treatment of hypercortisolism in the EU. It should also be noted that many assays for cortisol significantly cross-react with compound S, 11-deoxycortisol, and this may falsely elevate apparent cortisol levels in patients on metyrapone unless new mass spectrometric assays are used.

Ketoconazole inhibits the first step in cortisol biosynthesis (side-chain cleavage) and, to a lesser extent, the conversion of 11-deoxycortisol to cortisol; it is an even more potent inhibitor of C17-20 desmolase, decreasing androstenedione and testosterone production. Ketoconazole has probably been the most widely prescribed agent used to inhibit adrenal cortisol synthesis, but in July 2013 the FDA and European Medicines Agency issued a safety warning about the use of ketoconazole for the treatment of fungal infection because of its hepatotoxicity. Moreover, as it has never been subjected to rigorous clinical trial, its optimal use has not been fully defined as yet.

Metyrapone is an 11-β-hydroxylase (P450c11) inhibitor that blocks the final step in cortisol biosynthesis. By decreasing cortisol levels, ACTH increases and stimulates adrenal androgen production, which can cause an increase in hirsutism in women, thus precluding the use of metyrapone as the first-line drug for women requiring long-term control of hypercortisolism. Like ketoconazole, its use for treatment of CD has been an off-label use, although this is currently changing. The rate of control of hypercortisolism varies according to different studies, in part because all such studies were small and differed in patient characteristics, previous treatment, medication type and doses, length of follow-up, and criteria used to define disease control. However, it does seem to become effective within hours. Valassi *et al.* demonstrated urinary-free cortisol (UFC) normalisation in 57% of patients treated with metyrapone alone, of whom 46% also showed clinical control of the disease [32].

Etomidate is an intravenous substituted imidazole anaesthetic drug that blocks 11-β-hydroxylation of deoxycortisol to produce cortisol. It may be useful in hospitalised patients, when infused intravenously at a low dose [33]. It is the only available agent for patients unable to take medication by mouth and can be a useful therapy for severe hypercortisolaemia in patients intolerant of or unable to take oral medication.

Mitotane, an adrenocorticolytic drug used primarily for the treatment of adrenal carcinoma, can also be used to achieve medical adrenalectomy with or without pituitary irradiation in patients with CD [34]. However, its harmful secondary effects and the delayed onset of action constitute major limitations of its use in the treatment of CD [35]. Gastrointestinal (47%) and neurological intolerance (30%) are the most frequent adverse effects reported and, with the exception

Table 2. Indications for medical therapy in Cushing's disease.

Type	Medication	Mechanism of action/site of action	Dosage	Efficacy	Administration	Side-effects	Contraindications
Adrenal steroidogenesis inhibitors	Ketoconazole	11β-hydroxylase inhibitor	200 – 400 mg tds	30 to 90% in small studies	Oral	Gynaecomastia, alopecia, reduced libido, hepatoxicity, rahes, gastro-intestinal upset	Liver impairment, pregnancy,breast feeding,porphyria
	Metyrapone	11β-hydroxyl and 17,20-lyase inhibitor	250 – 1000 mg tds-qds, max 6 g/day	Up to 80%	Oral	Gastro-intestinal upset, acne, hirsutism, hypo or hypertension, oedema, hypokalae-mia, sedation	Pregnancy,breast feeding,severe liver impairment,porphyria
	Etomidate	11β-hydroxylase inhibitor	0.01 – 0.5 mg/kg/h	Rapid control of hypercortisolism	Intravenous infusion	Sedation, nausea and vomiting, temporary uncontrolled muscle movements,rash, oedema	Pregnancy,breast feeding,porphyria
	Mitotane	Adrenolytic, inhibition of cortisol synthesis and side chain cleavage; 11b-hydroxylase and 18 hydroxylase	500 – 1000 mgtds-qds, gradually increased from 500 – 100 mg/day to max 6 g/day	60 to 80%	Oral	Gastro-intestinal upset, deranged LFTs and TFTs, hype-cholesterolemia, ataxia, confusion, teratogenic, ortho-static hypotension	Pregnancy,breast feeding,stage 4 – 5 renal failure, severe liver impairment
ACTH inhibitors	Cabergoline	Dopamine receptor subtype 2 agonist	1 – 7 mg/week	Normalisation of 24-h UFC in 25 to 35% of patients in relatively small studies	Oral	Postural hypotension, nausea, sedation, hallucinations, oedema, depression, possibility of heart valve sclerosis	Porphyria, pregnancy, hyper-sensibility to ergot derivates, valvulopathy
	Pasireotide	Somatostatin analogue (binds to receptor subtypes 1, 2, 3 and 5)	600 – 900 µg twice daily	Normalisation of 24-h UFC in up to 26% of the patients; 9 to 43% decrease in tumour volume	Subcutaneous injection	Hyperglycemia, colelithiasis, diarrhoea, headache, sinus bradycardia	Severe liver impairment, avoid poorly controlled diabetes
Cortisol recep-tor blocker	Mifepristone	Glucocorticoid recep-tor antagonism	300 – 1200 mg daily	Improvement in hyperglycemia associated with Cushing disease (diabetes group: 60% and hypertension group: 38%)	Oral	Nausea, vomiting, dizziness, hypokale-mia, adrenal insufficiency, headache, arthralgia, increased TSH, decreased HDL, endometrial hyperplasia, rash, oedema	Severe asthma, porphyria, renal failures, severe liver impairment, breast feeding

HDL: High-density lipoprotein; TSH: Thyroid-stimulating hormone.

of those with persistent hypercortisolism, almost all patients required concomitant hydrocortisone replacement [34]. Moreover, it was shown that mitotane can have direct pituitary effects on corticotroph cells. This drug was demonstrated to affect cell viability and function of human and mouse ACTH-secreting pituitary adenoma cells. This inhibitory effect is caused by mitotane at concentrations corresponding to the adrenocortical carcinoma therapeutic window [36]. The promising *in vitro* data showing efficacy of mitotane in CD at the doses used for adrenocortical carcinoma must be confirmed in clinical trials, balancing the efficacy of reducing ACTH secretion with the development of side effects. The therapeutic window for patients without an adrenocortical carcinoma is likely to be much less.

3.2 ACTH inhibitors

Contrary to commonly-used medical therapies that do not target the pituitary tumour, *dopamine agonists* are potentially attractive agents for the treatment of CD because they act directly on the tumour and may have the potential for decreasing glucose intolerance and diabetes [5]. Indeed, functional dopamine receptor subtype 2 (DR2) is expressed in approximately 80% of corticotroph pituitary adenomas [37]. However, the results on the efficacy of dopamine agonist therapy in CD have been variable, and few patients experienced sustained improvement [38]. Bromocritine was shown to cause an acute decrease in ACTH, but this effect is not sustained over time with repeated dosing, and only few relatively small studies have examined the efficacy of cabergoline in patients with CD. In the study that includes the largest number of patients with CD treated with cabergoline (n=30), there was a normalisation of the 24-h UFC in 30% of the patients who were followed for up to 60 months, at a mean dose of 2.1 mg/week [39]. In patients with CD this drug is used at higher doses (up to 7 mg/week) than those used in patients with hyperprolactinaemia. This may raise some concerns regarding its long-term safety with regard to valvular heart disease. However, the dose is still significantly lower than those used for patients with Parkinson's disease, and the current data do not suggest a significant risk in this case [40]. The relatively low cost of cabergoline and its benign side-effect profile make it an attractive candidate for drug trials in patients with CD and a suitable therapeutic alternative during pregnancy [41].

Somatostatin analogues, and particularly the new multiligand somatostatin analogue, pasireotide, have been demonstrated to inhibit ACTH release in human corticotroph cells through interaction with type 5 somatostatin receptors [5,42,43]. As we will see in more detail below, these drugs can represent an alternative medical approach for CD. Pasireotide injection has been approved in a number of countries for the treatment of patients with CD for whom surgery has been unsuccessful or who are not surgical candidates [29]. It is now available in Europe and in the US and Canada. The major secondary effect associated to pasireotide therapy is the development of hyperglycaemia. Although its efficacy was comprehensively demonstrated by clinical trials, it presents a significantly higher cost compared to non-FDA approved drugs.

Retinoic acid (RA), used to treat various types of cancer, is a natural and synthetic derivative of vitamin A that regulates diverse cellular growth and differentiation programmes, and cell survival and death [44]. Its anti-proliferative and ACTH inhibitory effects have been demonstrated both *in vitro* and in experimental animals. In *in vitro* studies of AtT-20 pituitary ACTH-secreting tumour cells, RA was shown to causes an inhibition of ACTH secretion [45] seemingly restricted to ACTH-secreting tumour cells, since in rat normal pituitary cells neither ACTH, prolactin nor GH are affected by the treatment. Furthermore, RA inhibits cell proliferation and induces apoptosis in ACTH-secreting tumour cells [45]. The *in vivo* RA effects paralleled the *in vitro* effects [45]. A randomised study using RA in dogs with CD corroborated these findings [46]. Dogs were treated with RA (n = 22 dogs) or ketoconazole (n = 20 dogs) for a period of 180 days. In the ketoconazole group there were no significant changes in ACTH or α-MSH at any time studied. In contrast, in the RA treated dogs, there was a significant reduction in plasma ACTH and α-MSH over time. The survival time after initiation of treatment was significantly longer in the retinoic acid group compared with the ketoconazole group. Moreover, in the ketoconazole group, more than 50% of the animals died before completing the treatment, usually from complications of the glucocorticoid excess [46]. No adverse events (AEs) with RA were recorded, except for one case of footpad hyperkeratosis. More recently, a prospective multicentre study was the first to show a marked and prolonged decrease in UFC in three of seven CD patients, and ameliorated symptoms of hypercortisolism in five of them [47], supporting the possibility of RA representing a novel therapeutic approach to CD. In this series, albeit small, RA showed considerable potential as an ACTH secretion-restraining agent. Indeed, five of seven patients exhibited a clear-cut decrease in UFC excretion that led to normalisation in three cases. Furthermore, clinical features of hypercortisolism, in particular glycaemic control and body weight, were ameliorated. However, there remain doubts as to whether this will ever be a useful agent in this condition.

3.3 Cortisol receptor blocker

Mifepristone (previously known as RU-486) is an anti-progestational drug that is best known as an abortifacient. However, at much higher doses, it also acts as a GR antagonist. Apart from treatment of CD patients with hyperglyceamia, the role of mifepristone is not clear (as we will in more detail below) [31], but it may also be a reasonable short-term intervention for patients with an acute hypercortisolaemic crisis, such as cortisol-induced psychosis. This is an expensive drug with some particular pitfalls, such as a risk of mifepristone-induced hypokalaemia, a lack of available biological parameters of follow-up, and anti-progestin effects.

Several other different pharmacological approaches have been explored. Experimental studies demonstrated that PPARγ agonists blunted ACTH secretion in rodents with tumoral corticotroph implants [48]. Therefore, PPARγ agonists were hypothesised to reduce ACTH secretion via their anti-proliferative and pro-apoptotic effect; indeed, tumour growth was markedly blunted in rodents treated with rosiglitazone. These promising results, however, did not transfer to humans: pioglitazone or rosiglitazone administration in patients with CD failed to reveal consistent reductions in UFC, plasma ACTH or serum cortisol levels, possibly a consequence of the low proliferative potential of human ACTH-secreting pituitary tumours [49]. In addition, a potential concern for the long-term use of rosiglitazone in CD is its pro-osteoporotic effects, already observed in diabetic patients [50]. Temozolomide, an orally administrated second-generation alkylating chemotherapeutic agent, could be an efficient drug in cases of aggressive CD [51]. Temozolomide has shown promise as monotherapy [52], and in combination with pasireotide [53], as a treatment for aggressive pituitary adenomas and carcinomas, and may represent another viable treatment option targeting aggressive corticotroph adenomas refractory to surgery, RT, or other medical treatments. In tumours that responded to temozolomide, a prompt reduction in ACTH, chiasmaticde compression and reduced mass effects were observed [54].

4. Current research goals

CD is an excellent example of a situation where effective therapy is essential, but where the balance of risk and benefit must be carefully judged. In the case of CD, the re-evaluation of available drugs is still a valuable research goal, requiring proof of the effectiveness and applicability of currently available therapies. This 'recycling' of old drugs can bypass the limitations associated with side effects. Also, clinical trials evaluating these drugs should be designed with the highest quality standards, which in general has not always been the case to date.

Additionally, as part of all evolutionary processes, another important goal is to find new targets and therapeutic agents to test. The current research goals should identify and evaluate the efficacy of medical therapy in patients with severe biochemical disturbances (e.g., hypokalaemia), immunosuppression and/or mental instability, who may need immediate and life-saving cortisol-lowering therapy. Moreover, patients with CD are at risk of infection and cardiovascular events (both ischaemic and thromboembolic), and an evaluation before surgery is warranted to identify patients at high risk of perioperative complications and to optimise their overall health status before the operation. Also, an earlier diagnosis of patients with a high risk of post-surgical recurrence is important. Clinico-pathological studies have begun correlating the behavioural characteristics of pituitary tumours with histopathological and immunohistochemical features [55]. For CD specifically, the absence of peritumoral Crooke's change may be a predictor of recurrence after successful surgical treatment [56]. The absence of Crooke's changes indicates a lack of suppression, indicating that corticotroph cells may have some intrinsic abnormality predisposing to adenomatous islands and recurrence [56]. Overall, invasive pituitary adenomas tend to have a higher proliferative rate and immunopositivity for p53 [55]. Further assessment is required to characterise the histological features that may help predict long-term outcome and recurrence.

5. Scientific rationale

5.1 Ketoconazole

Ketoconazole, an imidazole derivative, is an antifungal agent that, in higher dosages, reduces adrenal steroid production via inhibition of multiple steroidogenic enzymes. This drug represents a racemic mixture of the cis enantiomers (-)-(2S,4R) and (+)-(2R,4S). One enantiomer of ketoconazole (2S, 4R) has been shown to be responsible for virtually all of the cortisol synthesis inhibitory activity present in the racemate. The industrial preparation of enantiomerically pure antifungal drugs with a high antifungal activity allows for the same therapeutic effectiveness using lower dosages than those required for racemic ketoconazole [57].

Apart from adrenal blocking effects, ketoconazole may also have direct effects on corticotrophic tumour cells of patients with CD. Stalla *et al.* [58] have shown that ketoconazole inhibits both basal and stimulated ACTH production by rat anterior pituitary cells. In addition, ketoconazole inhibited cell growth, in part by the induction of apoptosis. These effects may explain the absence of a compensatory rise in ACTH levels in patients with CD during prolonged treatment with ketoconazole [59], which contrasts with the observed ACTH increase during chronic treatment with metyrapone [60].

The major side effect of ketoconazole is hepatotoxicity, and liver function should be monitored carefully during treatment. In addition, ketoconazole can cause hypogonadism in men and gastrointestinal complaints [61]. It should not be administered with proton pump inhibitors as it requires gastric acidity for absorption.

5.2 LCI699

LCI699 is an 18-hydroxylase (aldosterone synthase) inhibitor and was first characterised as an aldosterone biosynthesis inhibitor for primary aldosteronism and essential hypertension. It also inhibits 11β-hydroxylase (CYP11B1) in a similar manner to the R-enantiomer of fadrozole (FAD286); this blocks the hydroxylation of deoxycortisol to cortisol as well as CYP11B2, thus blocking the conversion of deoxycorticosterone to corticosterone. As predicted from its mode of action, LCI699 treatment was associated with falls in circulating cortisol and aldosterone, and a rise in testosterone (in women), ACTH, 11-DOC and 11-deoxycorticosterone, the latter by 18-fold. The reduction in serum aldosterone was accompanied by a fall in circulating renin, presumably due to a rise in mineralacorticoids, and hypokalaemia was documented as

a drug-related adverse reaction. Taken together, these results suggest that the mineralocorticoid action of 11-deoxycorticosterone is sufficient to overcome any benefit from the inhibition of aldosterone synthase.

The dose of LCI699 required to match the benefit of the aldosterone antagonist eplerenone has been shown to induce hypoadrenalism, and this ability to inhibit cortisol secretion was transformed from a potential adverse reaction to a treatment goal. Currently, LCI699 offers a hope for managing patients with CD. Having established the ability of LCI699 to safely and effectively reduce cortisol levels in CD, the next step for researchers is to conduct a follow-up trial that enrols a greater number of patients, with longer treatment duration. Despite the similarities in the action mechanisms of LCI699 and metyrapone [60], LCI699 is a more potent drug (*in vitro* IC_{50} of 7.5 nM vs 2.5 nM for metyrapone) and appears to be effective at doses around 100 times lower [62]. In addition, LCI699 has a longer half-life [30], allowing a more convenient dosage schedule. Common AEs are gastrointestinal disturbance, including nausea and diarrhoea. And, as could be anticipated with such a potent drug, some patients had clinical effects that are consistent with adrenal insufficiency and/or steroid withdrawal (moderate fatigue, mild nausea, dizziness, mild muscle spasms and moderate hypotension), with these being effectively managed by a dose reduction.

The initial reports of LCI699 explored its aldosterone synthase (CYP11B2) inhibitory action, at doses of up to 2 mg/d in patients with primary hyperaldosteronism and essential hypertension, and demonstrated correction of hypokalaemia and hypertension [63,64]. The induced hypoadrenalism precluded the use of this drug, but unleashed the development of clinical trials evaluating its efficacy and safety in the treatment of CD.

5.3 Mifepristone

Mifepristone, a substituted 19-nor steroid compound, is the first and currently only available GR antagonist treatment [65]. Other GRs are currently being evaluated, but no clinical trial has been published to date [66]. Mifepristone blocks the action of cortisol by binding to the GR (type II) and blocks progesterone at the progesterone receptor. Steroid hormone receptors belong to the superfamily of nuclear receptors and are hormone-dependent transcription factors, positively or negatively regulating a large set of genes. They consist of a carboxy-terminal ligand-binding domain, a central DNA-binding domain that interacts with specific DNA sequences on target genes, and an N-terminal hypervariable region [67]. The GR is a member of the steroid hormone receptor subfamily of nuclear receptors, ubiquitously expressed in almost all human tissues and organs. At the cellular level, the GR, an intracellular receptor protein, mediates the actions of glucocorticoids. Many responses have been reported to occur within seconds or minutes after exposure to glucocorticoids. Following steroid binding, receptors undergo a conformational change that is probably crucial for receptor interaction with cellular targets. Mifepristone, a receptor antagonist, reversibly maintains the receptor in an inappropriate conformation. Mifepristone binds to the human GR with an affinity 3-4 times higher than that of dexamethasone and about 18 times higher than that of cortisol [68].

Mifepristone also has a strong anti-progestin activity, and a weak anti-androgen activity. Its affinity for the progesterone receptor is more than twice that of progesterone, and affinity for the androgen receptor is less than one third that of testosterone. It does not bind to the oestrogen receptor or the mineralocorticoid receptor. Following oral administration of a single dose of 400 - 600 mg, mifepristone is rapidly absorbed, with a peak plasma concentration occurring approximately 90 min after ingestion. Notably, in the absence of progesterone or cortisol, mifepristone can also act as a weak agonist [66]. Mifepristone appears as a rapidly effective drug in controlling signs of hypercortisolism.

As mentioned, mifepristone has the drawback of important secondary effects and limitations. About one third of the patients treated with mifepristone present with hypokalaemia [61]: as mifepristone blocks only glucocorticoid action, the mineralocorticoid activity of cortisol excess is not affected and can lead to hypokalaemia. The same mechanism can induce increased blood pressure levels, despite an improvement of the clinical signs of hypercortisolism. Also, during the treatment with this GR blocker, the levels of ACTH and cortisol increase, making it difficult to determine the efficacy of the drug. Management of adrenal deficiency is also difficult: hydrocortisone is not effective and should be given in parallel with mifepristone withdrawal. Due to its anti-progestin effects, this drug can also induce endometrial hyperplasia, and a pelvic ultrasonography should be performed yearly in every pre-menopausal woman.

5.4 Pasireotide

Soon after its discovery in 1972, somatostatin was known to be a major regulator of GH release from the pituitary and was therefore of interest for the potential treatment of acromegaly [69]. The production of synthetic somatostatin analogues with a significantly longer half-life was a major step forward in the treatment of acromegaly and gastro-enteropancreatic neuroendocrine tumours. Another important step was the discovery of the fivr somatostatin receptor subtypes. Whereas growth-hormone producing adenomas generally express high levels of type 2 somatostatin receptor (sst2), other adenomas, such as corticotroph adenomas, not only express considerably lower levels of sst2 but also its expression of sst2 is under negative control by glucocorticoids, which can be internalised in the hypercortisolaemic state. Pre-incubation with dexamethasone decreases the expression of sst2 in corticotrophcells [70]. The type 5 somatostatin receptor (sst5) seems more resistant to the suppressive effect of glucocorticoids, and is expressed at highest level in corticotrophadenomas [70]. More recently, sst5 was found to play an especially crucial role in regulating ACTH release in these cells and that

sst5-targeting agonists were more effective than sst2-agonists in inhibiting ACTH release [70].

Pasireotide (SOM230) is a multi-ligand SS-analogue with high binding affinity for sst2 and sst5, with IC_{50} values of 1.0 and 0.16 nM, respectively [71]. Its binding profile, which includes high sst5-affinity, makes it a promising new drug in the treatment of CD. In preclinical studies, pasireotide demonstrated inhibition of ACTH secretion in cultured human corticotroph adenomas [42,72] and in murine AtT20 cells [72,73]. Furthermore, consistently with the model of glucocorticoid-mediated down-regulation of sstr2, pre-treatment of AtT20 cells with dexamethasone abolished the inhibitory effects of octreotide on ACTH secretion, but had no effect on the inhibitory action of pasireotide or somatostatin-14 [70]. Somatostatin analogues may also confer an indirect anti-proliferative effect through inhibition of angiogenesis (down-regulation of vascular endothelial growth factor), which limits tumour growth [74], and by reducing levels of growth factors and trophic hormones [74].

Adenomas from patients with normalised preoperative UFC excretion had a 10-fold higher sst2 mRNA expression compared with adenomas from patients with elevated preoperative UFC excretion. Moreover, a recently published study by de Bruin *et al.* [75] showed that GR-antagonising therapy with mifepristone was able to induce sst2 expression *in vivo* in ACTH secreting neuroendocrine tumours, which was previously found to be undetectable with somatostatin receptor scintigraphy. However, whether sustained eucortisolaemia induced by medical therapy induces re-expression of biologically active sst2 protein in corticotroph adenomas and, if so, whether this increases the ACTH-lowering potential of sst2-preferring somatostatin analogues or pasireotide, remains to be established. Because somatostatin receptors have also been found in the normal adrenal cortex and in adrenocortical tumours, pasireotide could have a potential adrenal-directed effect in addition to its pituitary-directed effect. However, at the moment there is no clear evidence of a direct effect of somatostatin and its analogues at the adrenal level. Further studies must be carried out on the role of these receptors in the pathophysiology of the adrenal cortex.

The major drawback associated with treatment with pasireotide is the development of hyperglycaemia, but secondary effects such as cholelithiasis, diarrhoea, headache and sinus bradycardia have also been reported. Another potential problem might be any direct effects of pasireotide therapy on GH and insulin growth factor 1 (IGF-1levels) in CD patients. It is well known that pasireotide, via sst5 receptor activation, directly decreases GH release and hence IGF-1 levels in patients with acromegaly. In patients with CD, sustained hypercortisolism by itself causes a state of relative growth-hormone deficiency and therefore these patients may be at greater risk of becoming GH-deficient. Current and future clinical studies with pasireotide in CD patients should therefore include careful investigation of the effects on the GH/IGF-1 axis.

6. Competitive environment

6.1 Ketoconazole

Most of the retrospective studies evaluating the efficacy of ketoconazole in CD were small, with only two studies enrolling more than 10 patients each, only 100 patients being studied in total [59,76], and limited patient follow-up. The benefit/risk balance for using ketoconazole in CD is therefore difficult to determine (Table 3).

A recent retrospective multi-centre study provided important insights on the use of ketoconazole as a sole medical agent for the treatment of hypercortisolaemia. This study included 200 patients with CD, the largest number ever reported as being treated with this agent, observed over a 17-year period in 14 French tertiary centres. When used before TSS in 40 patients for a mean period of 4 months, ketoconazole resulted in controlled UFC in 49% of patients, partial control in 36%, and no control in 15%. Clinical improvements in diabetes, hypertension and hypokalaemia were noted despite the short duration of therapy with a mean dose of 585 mg/d. When considering a longer duration of therapy of 160 patients, treated for a mean of 24 months as either primary or secondary therapy, 50% were controlled, 23% were partially controlled and 27% were not controlled. These data suggest that around one-third of the patients on long-term follow-up were inadequately treated, and that other medical therapies or other strategies should have been considered. Approximately 20% of patients had to stop treatment due to poor tolerability. Mild and major increases in liver enzymes were observed in 13.5% and 2.5% of patients, respectively. No fatal hepatitis was observed [77].

Ketoconazole, the (2S,4R) enantiomer contained in the racemate of ketoconazole, is in Phase III clinical trials at Cortendo for the treatment of endogenous Cushing's syndrome. The company had also been developing the drug candidate for the treatment of type 2 diabetes; however, no recent development has been reported for this research.

6.2 LCI699

In a recent proof-of-concept study, adult patients with moderate-to-severe CD ([UFC levels ≥ 1.5 x ULN [upper limit of normal]) received oral LCI699 for 10 weeks. LCI699 was initiated at 4 mg/d in two equal doses; the dose was escalated every 14 days to 10, 20, 40 and 100 mg/d until UFC normalised, whereupon the dose was maintained until treatment ended (day 70). The primary end point was UFC ≤ ULN or a ≥ 50%decrease from baseline at day 70. Twelve patients were enrolled and completed the study. Baseline UFC ranged over 1.6 – 17.0 x ULN. All 12 patients achieved UFC ≤ ULN or a ≥ 50% decrease from baseline at day 70; 11 (92%) had normal UFC levels at that time. After treatment discontinuation (day 84), UFC was ≤ ULN in 10 patients. Mean 11-deoxycortisol, 11-deoxycorticosterone, and adrenocorticotropic hormone levels increased during treatment and

Table 3. Competitive environment.

Compound	Company	Chemical name	Chemical Structure	Indication	Stage of development	Mechanism of action
ßCOR-003 (CORT-001; DIO-902; Normocort), a single 2S,4R enantiomer of ketoconazole	Cortendo	Piperazine, 1-acetyl-4-[4-[[2-(2,4-dichlorophenyl)-2-(1H-imidazol-1-ylmethyl)-1,3-dioxolan-4-yl]methoxy. phenyl]-, cis- [CAS]		Safety and efficacy of COR-003 in the treatment of endogenous Cushing's syndrome	Phase III	Glucocorticoid antagonist; corticosteroid synthesis inhibitor
LCI699	Novartis	(R)-4-(6,7-dihydro-5H-pyrrolo [1,2-c]imidazol-5-yl)-3-fluorobenzonitrile		Long term the safety and efficacy of LCI-699 in the treatment of CD	Phase III	Aldosterone synthase inhibitor
Pasireotide (SOM-230)	Novartis	Cyclo((4R)-4-(2-aminoethylcarbamoyloxy)-L-prolyl-L-phenylglycyl-D-tryptophyl-L-lysyl-4-O-benzyl-L-tyrosyl-L-phenylalanyl-)		Safety and efficacy of pasireotidesc in the treatment of CD; 2 treatment arms with starting dose of 10 mg/month or 30 mg/month.	Phase II/III	Somatostatin receptor 1, 2, 3 and 5 agonist

ACTH: Adrenocorticotrophin; CD: Cushing's disease.

Table 3. Competitive environment.

Compound	Company	Chemical name	Chemical Structure	Indication	Stage of development	Mechanism of action
Pasireotide LAR	Novartis	Cyclo((4R)-4-(2-aminoethylcarbamoyloxy)-L-prolyl-L-phenylglycyl-D-tryptophyl-L-lysyl-4-O-benzyl-L-tyrosyl-L-phenylalanyl-)		Safety and efficacy of pasireotide LAR 60mg in the treatment of. neuroendocrine tumours of the pancreas or duodenum, pituitary glands, Nelson syndrome or ectopic-ACTH-secreting tumours	Phase II	Somatostatin receptor 1, 2, 3 and 5 agonist
Mifepristone (C-1073)	Corcept Therapeutics	Butanoic acid, 4-[[1-[[(4-aminobutyl)amino] carbonyl]-3-methylbutyl] amino]-4		Safety, pharmacokinetics and dynamics of mifepristone in the treatment of children with refractory CD	Phase III	Glucocorticoid antagonist
HRA-052015 (Mifedren)	HRA Pharma	Estra-4,9-dien-3-one, 11-[4-(dimethylamino)phenyl]-17-hydroxy-17-(1-propynyl)-, (11ß,17ß)- [CAS]		Safety and efficacy of HRA-052015 in the treatment of CD due to ectopic ACTH secretion	Phase III	Glucocorticoid antagonist

ACTH: Adrenocorticotrophin; CD: Cushing's disease.

declined after discontinuation. Mean systolic and diastolic blood pressure decreased from baseline by 10.0 and 6.0 mmHg, respectively. LCI699 was generally well tolerated; most AEs were mild or moderate. The most common AEs included fatigue (7/12), nausea (5/12) and headache (3/12). No serious drug-related AEs were reported [62]. Based on these promising results, the results of larger Phase III trials currently underway are anticipated.

6.3 Mifepristone

The initial information documenting the efficacy of mifepristone for use in Cushing's syndrome came from case reports. In 1985, a patient with Cushing's syndrome secondary to ectopic ACTH secretion was treated with mifepristone [78]. Treatment resulted in resolution of clinical effects of Cushing's syndrome, redistribution of fat and improvement in hyperglycaemia and hypertension. However, its approval by the FDA was only obtained many years later, after an open-label, 24-week, multi-centre clinical study evaluated safety and efficacy of mifepristone for the treatment of Cushing's syndrome [79]. The SEISMIC study enrolled 50 patients from 17 centres with continued biochemical and clinical evidence of hypercortisolaemia after failing multimodality therapy, largely surgery and/or RT. Inclusion criteria were the concomitant presence of type 2 diabetes, impaired glucose tolerance or hypertension with at least two other signs or symptoms of Cushing's syndrome. Forty-three of the patients had CD, 4 had ectopic ACTH tumours, and 3 had adrenal carcinomas. In the diabetes cohort, patients were required to have received stable anti-diabetic regimens before enrolment and the regimens could not be advanced during the study. Of the 25 patients in the diabetes cohort who received mifepristone for at least 30 days, 60% (15 patients) responded with a 25% or greater reduction in glucose AUC during standard glucose tolerance testing ($p < 0.0001$). Eighteen of the patients (72%) in the diabetes cohort had at least a 25% reduction in glucose AUC or were able to reduce anti-diabetic therapy. Mean reduction in A1C was 1.1% from baseline ($p < 0.001$), and 6 of 12 patients with an A1C greater than 7% at baseline had an A1C of 6% or less by the end of the trial. Mean ± s.d. fasting plasma glucose levels were reduced from 1.49 ± 0.747 mg/mL at baseline to 1.047 ± 0.375 mg/mL at 24 weeks ($p < 0.003$). The doses of anti-diabetic drugs were reduced in 7 of 15 patients. Of the 12 patients receiving insulin, 5 were able to reduce their insulin doses by at least 50% [79]. In the hypertension cohort, 8 (38.1%) of 21 patients achieved at least a 5 mmHg reduction in diastolic blood pressure compared to baseline ($p < 0.05$). Two of these responders received spironolactone. When the change in blood pressure was evaluated in the hypertensive patients in both cohorts (diabetes plus hypertension cohort), 42.5% (17 of 40 patients) had a reduction in diastolic blood pressure of at least 5 mmHg and 27.5% were able to reduce their antihypertensive drug doses. However, mean systolic and diastolic blood pressures were not significantly changed [79].

As patients included in the SEISMIC study represented a specific subset of individuals, it is unclear how generalisable these data would be to other patients with CD. Furthermore, the lack of an appropriate biochemical marker means that, in clinical practice, dose titrations would need to be based on clinical signs alone. In SEISMIC, the most common AEs were nausea, fatigue, headache, decreased blood potassium, arthralgia, vomiting, endometrial thickening and peripheral oedema. Adrenal insufficiency was reported in two patients, and there were five other cases in which symptoms consistent with adrenal insufficiency led to administration of glucocorticoids. Hypokalaemia was common (44%), but was mostly mild to moderate. Low potassium levels were often associated with alkalosis and oedema, which were usually reversible with potassium replacement. An increase in endometrial thickness was observed in 38% of the females in the study. There were five cases of vaginal bleeding [80].

The development of a selective GR without anti-progestin effects could also represent an important step in the long-term treatment of women with CD. Phase III trials evaluating the safety, pharmacokinetics and dynamics of mifepristone are ongoing.

6.4 Pasireotide and Pasireotide LAR

Pasireotide is a medication that has been studied in a high-quality randomised double-blind clinical trial in patients with CD [81], and based on Phase III trial results, it has approved in the EU and US for the treatment of adult patients with CD for whom surgery has failed or is not an option.

In a Phase-II proof-of-concept open-label, short-term multicentre study, designed to assess the efficacy, safety and pharmacokinetics of pasireotide in patients with CD, subcutaneous pasireotide (600 mcg twice daily) was shown to reduce UFC levels in 76% of the patients. Moreover, after 15 days of treatment, mean 24-h UFC was normalised in 17% of patients [82]. These results demonstrated for the first time that pasireotide is promising as a pituitary-targeted medical treatment for CD. Indeed, this was reinforced during a randomised, double-blind multi-centre Phase-III study that included 162 patients with persistent or recurrent disease or who were ineligible for surgery [81]. Patients were randomised to receive pasireotide 600 mcg (n = 82) or 900 mcg (n = 80) subcutaneously twice daily for 12 months. Multiple UFC measurements were used to determine efficacy. After 3 months of treatment, patients with mean UFC of at least two times the upper limit of the normal range or higher than baseline were unblinded and their dose increased by 300 mcg. The proportion of patients at 6 months with UFC levels at or below the ULN without a prior dose increase was 12/82 patients (15%) in the 600 lg group and 21/80 patients (26%) in the 900 lg group. Decreases in UFC from baseline to months 6 and 12 were statistically significant in both treatment groups ($p < 0.001$). Overall, signs and symptoms

improved as mean UFC decreased. Interestingly, at 12 months, tumour volume was reduced by 9% and 44%, in the 600 and 900 mcg groups, respectively. This Phase III study is limited by the lack of a control group and an imbalance in baseline UFC levels between the two treatment groups, which may have had an effect on outcome. The patients had improvement in blood pressure, an average 6.7-kg decrease in weight and improvement in their physical appearance. Pasireotide was most effective in those with milder disease. Although efficacy is usually seen within weeks, here normalisation of cortisol levels was achieved after months of treatment. It is conceivable that this late effect might be due to the reactivation of different SSTR, especially SSTR2, in concomitance with cortisol decrease. It is noteworthy that short-term results are not always predictive of long-term outcomes. Several case reports also suggest that there is no loss of efficacy after long-term therapy with pasireotide [83-85], although more data are needed.

Although pasireotide has a generally tolerability profile similar to that of other somatostatin analogues, it is associated with a relatively high incidence of hyperglycaemia, requiring the addition or intensification of glucose-lowering medication in a substantial proportion of patients. In the Phase III study, 73% of patients experienced glucose metabolism changes reported as hyperglycaemia (40%), diabetes mellitus (18%) or increased HbA1c (11%) [81]. Two studies in healthy volunteers elucidated the mechanism of action of hyperglycaemia associated with pasireotide administration and identified the increase in glucose levels as secondary to a marked inhibition of insulin and incretin secretion (both glucagon-like peptide-1 [GLP-1] and gastric inhibitory polypeptide [GIP]), with minimal inhibition of glucagon secretion and no impact on insulin sensitivity. Incretin-based anti-hyperglycaemic agents such as dipeptidyl peptidase-4 (DPP-4) inhibitors (e.g., vildagliptin) and GLP-1 analogues (e.g., liraglutide), followed by insulin secretagogues (e.g., nateglinide), may be able to ameliorate the effects of pasireotide on glucose homeostasis [86]. Further research into hyperglycaemia following pasireotide treatment will help refine the optimal strategy in CD. Other adverse effects related with pasireotide treatment were hypocortisolism (8%) [81], prolonged QT interval and decreased heart rate [87].

Long-acting pasireotide formulations, with a monthly dose frequency, have been developed to reduce the number of injections from twice a day to once a month in order to enhance convenience and improve its effectiveness. The use of this formulation is currently being investigated in CD, and enrolment is complete.

7. Potential development issues

7.1 Combination therapy

An additional potential line of development could be the realisation of more trials exploring the effectiveness of combination therapy on CD. Combination therapy has two major advantages. First, when approaching desperately ill patients it may guarantee an efficient and rapid inhibition of cortisol production. Second, by combining different mechanisms to improve hypercortisolism, they may provide improved efficacy with fewer side effects as lower doses of each drug may be used.

In a study of 14 patients with CD, a combination of ketoconazole and cabergoline achieved 24-h UFC normalisation in 79% of the patients [88]. The addition of ketoconazole to cabergoline and vice versa was effective, with no difference regarding which agent was initiated first [88]. This prospective analysis divided 14 patients (f/m = 12/2; median age 52, range 33 – 70 years) into two groups: 6 patients initially treated with cabergoline for 4 – 6 months (rising from 0.5 – 1 mg/week up to 3.0 mg/week), after which ketoconazole was added (group A); and 8 patients first took ketoconazole alone for 4 – 6 months (rising from 200 mg/day to 600 mg/day), then cabergoline was added (group B). Patients were compared with 14 age-matched patients in prolonged remission after effective neurosurgery for CD. The combination therapy led to UFC normalisation in 79% of patients with no differences between the groups; only one patient failed to respond at all. Neither drug succeeded in controlling the disease when taken alone. Associating cabergoline with ketoconazole may represent an effective second-line treatment, achieving a satisfactory reduction in UFC levels and clinical improvement. Although the combined treatment lowered patients' LNSC levels, they remained higher than normal, indicating a persistent subclinical hypercortisolism; the implications of this finding need to be considered.

Feelders *et al.* [83] conducted an 80-day study where pasireotide was initially administered as monotherapy, and cabergoline and low-dose ketoconazole were sequentially added at 4 and 8 weeks, respectively, as normalisation of urinary cortisol levels was achieved. This approach achieved normalisation in 90% of patients. Pasireotide monotherapy normalised UFC in 5 out of 17 patients (29%). Cabergoline addition resulted in normalisation in another four patients (24%), and all other patients, except one, experienced a mean 48% reduction in urinary cortisol levels. The addition of ketoconazole resulted in normalisation of urinary cortisol in six additional patients (35%). In another study, the addition of ketoconazole to cabergoline also increased the proportion of patients who achieved normalisation of urinary cortisol [84]. In a small study of patients with ACTH-dependent Cushing syndrome, which included four patients with CD with severe hypercortisolism (24-h UFC > 832 µg/day; normal, 10 – 65 µg/day), low or normal 24-h UFC was achieved in all patients with triple combination therapy of ketoconazole, metyrapone and mitotane [85]. The patients underwent pituitary surgery 5 – 22 months after initiation of combination therapy. Three patients achieved remission postoperatively, and one patient needed to resume medical therapy when hypercortisolism recurred. Adverse effects were tolerable, consisting mainly of gastrointestinal discomfort and a significant rise in total cholesterol and γ-glutamyltransferase levels.

Although the remission rates in these small studies are very encouraging, none of the combination therapies is currently approved in patients with CD. There is clearly a need for further studies examining the potential of combination therapies in these patients.

There is also a scientific rationale for combining pituitary-directed agents for the management of CD. Corticotroph cells co-express both somatostatin and dopamine receptors, which work synergistically through membrane interaction or dimerisation [89,90]. Studies on different tumour models, including pituitary adenomas, revealed a higher potency of BIM-23A779, BIM-23A760 and BIM-23A781 (chimeric molecules containing both SST and dopamine structural elements) in controlling tumour cell growth [91]. If the functional heterodimerisation of these receptors occurs *in vivo*, as has already been demonstrated *in vitro*, the combination of cabergoline plus pasireotide and/or other SSTR5-targeted somatostatin analogues and/or BIM23A760 (a chimeric dopamine-somatostatin receptor agonist) could be an exciting concept, increasing the medical options for these patients.

A prospective, Phase II trial assessing pasireotide, alone or in combination with cabergoline, in patients with CD is ongoing. In this study, 128 adult patients with persistent, recurrent or *de novo* CD will be enrolled and stratified into one of two groups: group 1 is pasireotide naive or with treatment discontinued ≥ 4 weeks before screening because of lack of efficacy; and, group 2 received maximum tolerated dose of pasireotide for ≥ 8 weeks, without biochemical control. The primary end point of the study is to evaluate the proportion of patients of group 1 with mean urinary free cortisol ≤ ULN at week 35 with pasireotide alone or in combination with cabergoline, and the proportion of patients of group 2 with mean urinary free cortisol ≤ ULN at week 17 with pasireotide in combination with cabergoline [92].

8. Conclusion

CD is a severe and complex entity which needs aggressive and rapid curative treatment due to its long-term sequelae. Unfortunately, current therapeutic options, even in the hands of the most specialised centres, do not achieve cure in a significant proportion of patients. Considering the effects of uncontrolled hypercortisolism on morbidity and mortality and the drawbacks of RT and bilateral adrenalectomy, there is a clear need for effective medical therapy for patients with CD with whom surgery is unsuccessful or not feasible. Therefore, the search for new medical effective tools remains open, and novel options are currently being explored.

9. Expert opinion

We now have a number of different agents available for the control of the clinical manifestations of hypercortisolism in varying situations. When rapid control of the hypercortisolaemia of CD is required, metyrapone is the drug of choice as it works within hours and is highly effective in some 75% of patients. However, in the long-term, it causes virilisation in women. Ketoconazole is a good second-line drug, is probably slower in onset and slightly less effective than metyrapone, but can also be used in combination. LCI699 is similar to metyrapone but more potent and longer-acting, and may in the future be a better alternative, but more data are required. Mifepristone is expensive and difficult to monitor, but can be used as oral medication in patients where metyrapone or ketoconazole are inappropriate: we do not see its extensive use. Where a rapid response is required and the patient is *in extremis*, intravenous etomidate can be life-saving.

It is difficult to prognosticate regarding future adrenostatic or adrenolytic drugs for CD. At present, the array we have available are moderately effective and often complementary, and the recent licensing for metyrapone and ketoconazole in particular is a welcome innovation for endocrinologists. LCI699 may have certain advantages over metyrapone, and if marketed might also provide competitive positioning with metyrapone such that overall costs will fall. At present, it seems unlikely that the availability of the specifically-active enantiomer of ketoconazole will offer any major advantages, although again such a new entry to the market may have cost benefits for patients and clinicians. It seems improbable, at present, that there will be any other new entrants to this arena in the immediate future.

For drugs working directly on the pituitary, cabergoline is occasionally effective, although escape may occur, but it is readily available. Pasireotide can be used in patients with mild CD, although its use is hampered by high cost and significant hyperglycaemia. We do not personally feel that this will play a major role in the treatment of CD, not least because of its high cost and diabetogenic potential. The real requirement is for an orally active drug effective in the majority of patients with CD with no major adverse effects, and none is currently foreseen. It is possible that combined dopamine-somatostatin analogues will be more useful than pasireotide, but it would seem that any somatostatin subtype 5 receptor agonist is going to be a potent inhibitor of incretins and insulin, which is a powerful disincentive to their use.

As most corticotroph tumours respond to CRH with the release of ACTH, potent CRH-antagonists may be a completely novel approach, and their development should be encouraged. As an alternative approach, botulinum toxin can be linked to a CRH ligand and therefore target corticotroph cells to block ACTH secretion and possibly also tumour cell viability. Such a development would be even more innovative, and we would be strongly supportive of pharmaceutical research and innovation in this area.

Declaration of interest

D Guelho has no relevant affiliations. AB Grossman has received lecture and consultancy fees from Novartis, Ipsen and HRA Pharma. The authors have no other relevant affiliations or

financial involvement with any organisation or entity with a financial interest in or financial conflict with the subject matter or materials discussed in the manuscript. This includes employment, consultancies, honoraria, stock ownership or options, expert testimony, grants or patents received or pending, or royalties.

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