

# North Thames Clinical Chemistry Audit & QA Group

## Meeting Report:

### Audit of Aldosterone and Renin Investigations 14<sup>th</sup> July 2004

#### Results of the Aldosterone & Renin Questionnaire

Dr Pat Kyd, SAS Aldosterone & Renin laboratory, St Mary's Hospital

See PowerPoint Presentation (Appendix 1)

#### Summary

- 1) 44 questionnaires were circulated to laboratories in the North Thames and Eastern Regions with a response rate of 52% (23)
- 2) Most laboratories refer their work (19/23 for renin and 20/23 for aldosterone). Three laboratories measure aldosterone locally and four laboratories measure renin locally.
- 3) 4 laboratories reported that they recommend that renin samples are sent on ice. Samples for plasma renin activity must not be sent on ice because of the potential to activate prorenin to form renin. Samples should not be chilled but sent to the lab at room temperature, separated and stored at  $-20^{\circ}\text{C}$  ASAP.
- 4) All laboratories reported that samples reach the laboratories in less than four hours.
- 5) Different reference ranges were quoted for aldosterone (4) and renin (5) even though only a total of 3 and 4 laboratories, respectively, measure these hormones.
- 6) Aldosterone and renin are requested in the following clinical situations:  
?primary hyperaldosteronism (23/23), electrolyte disturbances (20/23), Mineralocorticoid replacement (20/23), secondary hypoaldosteronism (19/23), pseudohypoaldosteronism (17/23), renal artery stenosis (11/23) and secondary hyperaldosteronism (8/23).
- 7) Most laboratories measure both renin and aldosterone, although some will only measure renin when monitoring mineralocorticoid replacement therapy. Most laboratories report the aldosterone/ratio ratios (ARR) if appropriate (19).
- 8) Most laboratories have written protocols (18) and most advise that spironolactone (19) and beta-blockers (18) should be stopped prior to sampling. Most laboratories use random aldosterone/renin ratios (18) with 5 using postural studies. All laboratories should be advising that B-blockers and spironolactone be stopped as false positive ARR with false negative renins are obtained. Calcium channel blockers, ACEI and diuretics may be less of a problem but should be stopped for two weeks if at all possible (may see false negative results). Drugs may be substituted with alpha-blockers which have no significant effect on the ARR.
- 9) There seems to be a lack of information supplied to the laboratory measuring aldosterone and renin which would be required for full interpretative comments. However laboratories generally passed on the information they had to the referral laboratory.
- 10) Most but not all laboratories (15/20) always receive clinical comments from the referral laboratory and those that receive comments report them to the clinicians.
- 11) After a positive screen 15/23 laboratories repeat the ARR off any interfering drugs and 11 laboratories request postural studies. No laboratory reported the use of fludrocortisone. 15 laboratories offer postural studies; however a variety of protocols were in use.

12) Most laboratories reported that adrenal vein localisation is only performed after the diagnosis of primary hyperaldosteronism (16). Patients are referred to a centre with experience for this procedure (16)

### **Clinical Biochemistry of Aldosterone & Renin**

Mr Mike Scanlon, SAS Aldosterone & Renin laboratory, St Mary's Hospital

See PowerPoint Presentation (Appendix 2)

The role of the renin-angiotensin-aldosterone system in blood pressure homeostasis and maintenance of sodium-potassium balance was described. The biochemical investigation of abnormalities of the system was described including the preparation of the patient, sample requirements, and the information to be sent with the request which is needed to interpret aldosterone and renin results. The causes and investigation of primary hyperaldosteronism were described. The use of the random aldosterone/renin ratio (ARR) was discussed. Postural tests are not useful, dynamic tests are infrequently used and urine aldosterone measurement is no longer offered as audit has shown it contributes little additional information. Other disturbances in the renin aldosterone pathway were described e.g. mineralocorticoid deficient states. The indications for laboratory measurement of renin and aldosterone were illustrated by case presentations. Indications include: the investigation of possible primary hyperaldosteronism (resistant hypertension with sodium retention and/or hypokalaemia), localisation of renal artery stenosis, diagnosis of renin-dependent hypertension, monitoring the adequacy of mineralocorticoid replacement in congenital adrenal hyperplasia and suspected hypoaldosteronism (low sodium and raised potassium).

### **Hyperaldosteronism and Hypertension: the Clinical View**

Prof Morris Brown, Clinical Pharmacology Unit, Addenbrooke's Hospital and University of Cambridge

See PowerPoint Presentation (Appendix 3)

Conn described the syndrome in 1953. The proband was a female with hypertension (BP176/104) with hypernatraemia, hypokalaemia and an alkalosis. She was found to have a large right adrenal adenoma. The textbook frequency of Conn's is 1% of patients with hypertension, but it is underdiagnosed. In 1990's the use of ARR by several groups suggested the prevalence of Conn's was much higher, in both HT clinic and primary-care populations. Richard Gordon published a detection rate of greater than 10%. However, there are flaws in his data as he used a selective hospital population. Prof Brown described his PHARst study, the Prevalence of **Primary Hyperaldosteronism** measured by **Aldosterone to Renin ratio** and **Spironolactone Testing**. If the ARR was found to be greater than 800, a CT of the adrenals was performed. If the ARR was between 400 and 800, the patient was given a trial of spironolactone and if the systolic blood pressure then dropped by >20 mmHg, the adrenals were scanned. This study showed that although a high aldosterone/renin ratio is common (14% had a ratio>800), Conn's is uncommon. (only 1 patient had an adrenal adenoma).

The data illustrated some of the drug effects on the ARR. The most marked effect is seen with beta-blockers which increase the ratio as they act by markedly suppressing renin with no effect

on aldosterone. Calcium blockers and diuretics make little difference to renin release. ACEI cause an increase in renin release brought about by low angiotensin (negative feedback). Drug issues are very important; however it is sometimes very difficult to remove patients from drugs, as it is important that their hypertensive control is not compromised.

It is vital to use appropriate cut-offs for ARR, which are method dependent. The cut-offs for the RIA method ( PRA in pmol/ml/hr) are quite different for those for the Nichols assay ( Active Renin in  $\mu\text{U/ml}$ ).

A suppressed renin in a patient on an ACEI is a good predictor of response to spironolactone. Thiazide diuretic therapy should not decrease plasma potassium by more than 0.1-0.2 mmol/L; if there is a greater fall in potassium, Conn's should be considered. Also, if sodium does not fall, Conn's should also be considered. In hypertension with low renin and low aldosterone, which responds to spironolactone therapy, consider a rarer syndrome such as AME, GRA or Liddle's. Gordon's syndrome is also a possibility, however, this is very rare. In this familial condition there is a low renin, low aldosterone and raised potassium : therapy includes a low potassium diet and a thiazide diuretic.

Investigations for secondary hypertension in drug resistant patients should include electrolytes, renin and urine VMA/catecholamines. Hypokalaemia needs to be treated before investigations start. Renin is the most important; aldosterone is not always required if the plasma renin is incompatible with primary hyperaldosteronism. In further investigation of primary hyperaldosteronism, an MRI or CT of adrenals should be performed (little to choose between); MRI is better for looking at tissue texture. To diagnose a unilateral adenoma there needs to be biochemical evidence (raised aldosterone, decreased renin), anatomical evidence (MRI or CT) and functional evidence- e.g. blood pressure response to spironolactone, and biochemical evidence of unilateral hypersecretion. If adrenalectomy is considered, adrenal vein sampling to localise aldosterone output is mandatory but is difficult and needs to be done at specialist centres. It is mostly performed by the laproscopic route (9/10). The right adrenal vein drains into the inferior vena cava which can only be accessed by a hook shaped catheter.

In summary: Classical primary hyperaldosteronism is present in <2% of hypertensives. Much more common is **normal aldosterone spironolactone sensitive hypertension (NASSH)** which may be part of the spectrum of low renin resistant hypertension. Most PHA can be suspected in patients selected because of hypertension resistant to several anti-hypertensive drugs from their plasma sodium, potassium and renin levels. Beta-blocker therapy may result in 'false positives'. It is much easier to diagnose PHA when patients are on ACEI and ARB therapy.

Prolonged treatment with spironolactone can cause problems e.g. gynaecomastia. An alternative drug is eplerenone which is a more selective aldosterone blocker without the side effects of spironolactone, but it is expensive. High dose amiloride is also very good.

### **Discussion Points**

1. The SAS lab often receives insufficient clinical information with the sample request, and it is appropriate to measure both aldosterone and renin
2. There is an increased use of the Nichol's Active Renin assay, but it needs to be more robust. The Nichol's assay is a very good assay however the correlation with the RIA method at the lower end is poor. An overlap between normals and Conn's was noted at the time it was first evaluated. However this was a couple of years ago, and there were

manufacturing production problems. Barts have noticed that there have been problems with the Active Renin assay – the standard does not cover the whole reference range. They also found it difficult to differentiate normal from low renin, which is essential for the diagnosis of primary hyperaldosteronism.

3. 80% cases of PHA are bilateral hyperplasia which may well respond to spironolactone.
4. Renin may be used to guide treatment in drug-resistant patients. These patients should be seen in specialist centres: renin investigations should not be sent from primary care. Patients who are resistant to three classes of drugs should be referred
5. If a patient has hypokalaemia whilst on ACE inhibitors, PHA should be considered.
6. When reviewing electrolyte results, there needs to be awareness of problems of pseudo-hyperkalaemia in primary care samples which may mask hypokalaemia
7. Low sodium diets may mask PHA and may result in normal potassium levels: patients must be maintained on a normal sodium intake prior to investigation.
8. More emphasis should be placed on plasma sodium. It is rare to see a sodium of less than 140 mmol/L in a PHA patient.
9. Hypertension clinics may measure renin to determine drug treatment. However, this is only feasible with a high throughput assay.
10. Postural studies are of no use, especially if the patient is on B blockers, and of historical interest only. CT scanning and a trial of spironolactone are more informative.
11. Afro-Caribbean's have lower plasma renin and aldosterone levels. This ethnic group requires a different data set and algorithm for interpretation of results
12. Plasma aldosterone upper limit of normal is probably 500 pmol/L ; the quoted 800pmol/L is very high. No Conn's case with proven adenoma has been reported with an aldosterone less than 400pmol/L.
13. There is currently no 'secondary hypertension register' but one is being set up by the British Hypertension Society. St Mary's has a database going back several years, but it relies on clinical details on request forms coming from the referring laboratories to make the diagnosis of PHA. Prof Brown and Dr Simon Thom ( St Mary's) hoped that the SAS labs would be closely involved in the creation and maintenance of this register.

## Standards/Guidelines

The draft standards/guidelines proposed at the meeting were accepted.

## Guidelines for Investigation of primary hyperaldosteronism

### 1. Who to screen

- a) hypertension with spontaneous or diuretic- induced hypokalemia especially in presence of a sodium  $\geq 140$
- b) hypertension refractory to 3 or more drugs
- c) young hypertensives
- d) hypertension in presence of an incidental adrenal adenoma

NB: Plasma potassium itself is not a good indicator of which patients need investigation: a significant proportion of patients with primary hyperaldosteronism are normokalaemic

### 2. How to screen

- a) first correct severe hypokalaemia ( plasma potassium should be not lower than 2.9 mmol/L) and ensure a normal salt intake. Potassium supplements, if given, should be stopped 24 hr prior to sampling
- b) measure both plasma aldosterone and plasma renin with the patient in a sitting position after resting for 15 minutes
- c) antihypertensive drugs may be continued with the exception of spironolactone, oestrogen ( stop for 6 weeks) and  $\beta$  blockers (stop for 2 weeks). An  $\alpha$  blocker such as doxazosin may be substituted.
- d) current drug information should be given on the request form, together with the statement that the patient is hypertensive and their recent plasma and urine electrolyte results
- e) samples should be sent to the lab at room temperature to arrive within 4 hours

### 3. The local laboratory

- a) The laboratory should have a protocol for the investigation of ? Conn's available to clinical users.
- b) Samples for renin should be separated within 4 hours of being drawn and the plasma frozen quickly. The sample must remain frozen until analysis. If the sample is sent to a referral lab, sample packing should ensure it remains frozen.
- c) The results from the referral laboratory, together with any interpretative comments, should be reported in full by the local hospital.

### 4. Interpretation of plasma renin and aldosterone results

Note that values for plasma aldosterone and renin ( activity or mass) are method and unit dependent. The following recommendations for the interpretation of aldosterone, renin and the aldosterone renin ratio apply only to results obtained by the SAS laboratories at St Mary's and Leeds, in the units quoted

- a) Both plasma renin and aldosterone should be measured. A renin result alone may be useful in excluding primary hyperaldosteronism if it is normal or high, but the value of aldosterone result alone is very limited.
- b) First consider plasma renin: in primary hyperaldosteronism it is suppressed or very low e.g.  $\leq 0.3$  pmol/ml/hr ( ref range 0.5-3.1).

- c) Plasma aldosterone may be raised or within the reference range ( 100-800 pmol/L) in primary hyperaldosteronism but must be  $\geq 250$ pmol/L
  - d) Providing the patient is known to be hypertensive, the aldosterone/renin ratio may be helpful. This ratio is less affected by variables such as intra-personal variation, drug therapy than its individual components and so can be used under random conditions. An ARR  $< 800$  usually excludes Conn's, whereas ARR  $> 2000$  makes it very likely.
  - e) If both renin and aldosterone are suppressed in a patient with severe hypertension , consider other sources of mineralocorticoid-like activity e.g. liquorice, carbenoxelone, cortisol excess, AME, 17  $\alpha$ -hydroxylase deficiency, Liddle's syndrome
5. Further investigations
- a) Repeat the plasma aldosterone, renin, and ARR after stopping any interfering drugs spironolactone ,  $\beta$  blocker, ACEI. It is essential that the patient is not on spironolactone ( false negative ARR ) or  $\beta$  blocker ( false positive ARR)
  - b) In primary hyperaldosteronism there is inappropriate potassium loss in the urine, demonstrated by a 24h output  $> 30$  mmol in presence of hypokalaemia. Urine potassium output is not helpful if the patient is normokalaemic.
  - c) Urine aldosterone measurement has been discontinued by SAS service.
  - d) The posture test adds little extra information to the screening test; it is not reliable in distinguishing unilateral adenoma from bilateral hyperplasia
  - e) Dynamic tests (saline infusion test, fludrocortisone suppression test, captopril test) may demonstrate the autonomy of adrenal aldosterone secretion but are not often employed, as they require hospitalisation of the patient, may not be safe, and may not be appropriate for all ethnic groups
6. Localisation of the source of aldosterone excess is required if surgery is considered:
- a) the diagnosis of primary hyperaldosteronism must first be established
  - b) adrenal imaging by CT scan or MRI ( possibly the more sensitive technique) should then be undertaken to try to locate an adenoma. Adenomas may be too small ( $<5$ mm) to be detected by imaging. The possibility that an observed adenoma is a non-secreting incidentaloma must be borne in mind.
  - c) Selective adrenal vein sampling is often an very useful tool to confirm or refute a unilateral source of excess aldosterone secretion. It is technically difficult to perform and should be conducted only in centres with radiologists who have experience in performing this technique.

### **Investigation of Mineralocorticoid deficiency and Fluid/electrolyte disorders**

1. Hypoaldosteronism should be considered in any case of unexplained hyperkalaemia with hyperchloraemic acidosis. Concomitant PRA measurement will distinguish whether hypoaldosteronism is primary (high PRA) or secondary to hyporeninaemia.
2. Measurement of PRA alone is appropriate when monitoring mineralocorticoid replacement e.g. CAH. Optimal replacement is associated with a PRA within the reference range. Aldosterone and PRA measurement has no role in the diagnosis of CAH or Addison's disease.

3. Consider pseudohypoaldosteronism as a cause of hyponatraemia, hyperkalaemia, renal salt wasting in infants in whom CAH has been excluded. End-organ resistance to aldosterone is associated with extremely high levels of both aldosterone and renin. Failure-to-thrive, recurrent chest infections and elevated sweat sodium may allow this condition to be mistaken for cystic fibrosis. Severe renal disease in infants may mimic pseudohypoaldosteronism.
4. The only form of secondary hyperaldosteronism in which measurement of aldosterone and renin is of value is Bartter's/ Gitelman's syndromes. Patients are normotensive with hypokalaemia and a hypochloraemic metabolic alkalosis, and may be hypomagnesaemic (Gitelman's). Vomiting, diarrhoea and diuretic/laxative abuse should first be excluded.

P Kyd/M Scanlon Jun 04

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