The Silent Diabetes Pandemic

Overcoming the current challenges in ketone testing

A striking statistic is that an estimated 4.2 million people die annually from Diabetes complications and it is anticipated it will be the seventh leading cause of death by 2030 according to the World Health Organisation (WHO)[1].

Type 2 diabetes mellitus (T2DM) has now reached the status of 'global pandemic', a phrase most of us are familiar with as a result global Covid-19 pandemic, but one we now know not to take for granted.

Diabetic Ketoacidosis (DKA)

Ketosis is a metabolic process that occurs when the body switches from glucose to predominantly fat metabolism for energy production, this happens when carbohydrate availability reaches low levels.

The metabolism of fatty acids in the liver results in the production of chemical by products known as ketone bodies or ketones [2]. Ketosis occurs when the body produces more ketones than the liver can process.

DKA is a serious complication of Type I Diabetes Mellitus (T1DM), however can also affect individuals with T2DM. The condition is linked to insulin deficiency and occurs when glucose levels are consistently high and insulin levels are severely low.

Due to this imbalance glucose builds up in the blood and the body responds by metabolising fat rather than glucose. DKA is usually one of the first indicators of T1DM.

Ketosis is not normally dangerous and is typical of ketogenic diets which are low in carbohydrates. Ketones however are poisonous when present in high levels leading to ketoacidosis, DKA for example if left untreated can cause damage to vital organs and in some instances may lead to a coma or death [3]. DKA is commonly triggered by an illness, infection or missing insulin treatments[4].

Clinical significance of D-3-Hydroxybutrate

There are three main ketones produced as a result of ketosis; D-3-Hydroxybutyrate, **Acetoacetate and Acetone**.

D-3-Hydroxybutyrate is the principal ketone accounting for 75% of total ketones in the body and is later catabolised into acetoacetate and then into acetone[5]. Due to the higher levels of D-3-Hydroxybutyrate, it is the more sensitive marker for the diagnosis of Diabetic ketosis.

Furthermore, D-3-Hydroxybutyrate also has a greater stability compared to either acetone or acetoacetate and is typically stable for 7 days at 4ºC making it clinically useful for the diagnosis and a patient's response to treatment.

The American Diabetes Association recommends testing for ketosis in diabetics when symptoms of ketoacidosis are present, i.e. when glucose levels are consistently elevated, during pregnancy and if experiencing any illness.

NICE also recommend monitoring ketones in patients with T1DM especially during periods of illness[6]

Additional Uses of D-3-Hydroxybutyrate

Ketosis occurs in a number of clinical conditions, therefore the detection of D-3-Hydroxybutyrate is of high importance.

- **Gestational diabetes** – Elevated ketone levels are common in pregnant women with gestational diabetes. If unchecked and untreated these ketone levels can have serious consequences for mother and baby.
- **Alcoholic ketoacidosis –** In Alcoholic ketoacidosis the ratio of D-3-Hydroxybutyrate to acetoacetate is more than twice that in DKA. Monitoring levels of D-3-Hydroxybutyrate is important when making a differential diagnosis between the two conditions.
- **Severe injury, illness or sepsis -** Many critically ill patients respond to starvation by metabolising fats and proteins, which leads to muscle wastage. Monitoring ketone levels in these patients following an overnight fast allows diagnosis of this problem. D-3 Hydroxybutyrate may be used to identify patients likely to benefit from nutritional support.
- **Ketotic hypoglycaemia -** This refers to any circumstance in which hypoglycaemia is accompanied with ketosis. It is also used to define recurrent episodes of hypoglycaemia with ketosis in young children. Delayed feeding is the most common cause among children with symptoms including vomiting, lethargy, unresponsiveness and even seizures. The detection of D-3 Hydroxybutyrate in the serum or plasma of infants may indicate kenotic hypoglycaemia.
- **Childhood epilepsy -** Regular monitoring of ketone levels may be beneficial in maintaining a ketogenic diet for the control of childhood epilepsy.

Limitations with existing methods used to measure ketones

Semi-quantitative, nitroprusside-based methods remain common for the detection of ketones in the blood and urine of diabetic patients. The nitroprusside method is available in both tablet and reagent test strip form where urine or blood is applied, and colour change compared to a colour chart. There are several limitations associated with **Nitroprusside methods**;

- Capable of detecting only acetone and acetoacetate, as such they lack sensitivity especially in early stages of DKA.
- The intensity of the colour change is subjective compared to quantitative methods which can be used to monitor ketosis to resolution and monitor improvements.
- Several medications including Valproic Acid and Vitamin C can interfere with the nitroprusside method leading to false positive results.
- False negative results are common as the method does not detect D-3-Hydroxybutyrate. As ketoacidosis improves and D-3Hydroxybutyrate is converted to acetoacetate the result becomes positive.
- D-3-Hydroxybutyrate is a more reliable indicator of ketosis and DKA due to its superior stability when compared to acetone and acetoacetate.

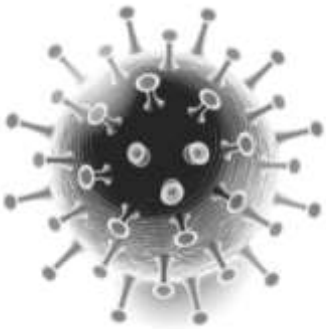
There are additional limitations associated with urine test strips. Blood D-3-Hydroxybutyrate testing is more effective than urine acetoacetate testing in reducing emergency department assessment, hospitalisation and time to recovery from DKA[7].

Table 1 below summarises the advantages of measuring ketones in blood via quantitative clinical chemistry assays compared to semi-quantitative urine test strips.

Table 1: Limitations with methods measuring Ketones in urine

Measuring Ketones in Blood	Measuring Ketones in Urine
Measures D-3-Hydroxybutyrtae, the predominant ketone in the blood.	Measures acetoacetate a poor indicator of overall blood ketone levels.
Provides a direct measurement of ketones in circulation.	Provides an indirect measurement of ketones in circulation and is therefore not as accurate.
Indicates current ketone levels allowing immediate medical intervention.	Indicates blood ketone levels in the previous 2-4 hours.
Unaffected by fluctuating fluid levels.	High fluid intake may dilute urine leading to false negatives. Low fluid levels or dehydration may lead to false positives.
Quantitative methods ran on clinical chemistry analysers are more reliable.	Semi-quantitative urine test strips are less reliable and subjective relying on the intensity of the colour change.

Diabetic Ketoacidosis in COVID-19?



Although individuals with diabetes appear to be at similar risk of contracting SARS-CoV-2 the virus that causes COVID-19 they are more likely to suffer severe consequences. There is increasing evidence to suggest that DKA is a common complication in those with Diabetes[8]. DKA was found to both increase the length of stay in hospital and mortality, meanwhile T1DM and T2DM alone had no effect on mortality but did increase hospital stay [9].

Randox D-3-Hydrooxybutyrate superior methodology

Blood ketone levels provide valuable information to clinicians allowing accurate and reliable identification of DKA. D-3-Hydroxybutyrate as the most abundant ketone in the body is a more sensitive and specific indicator of DKA compared to other available methods on the market. D-3-Hydoxybutyrate measurement enables earlier more reliable detection of ketosis and allows faster treatment.

D-3-Hydoxybutyrate demonstrates superior methodology when compared to other commercially available ketone detection tests. For example, the nitroprusside method used in semi-quantitative dipstick tests only detects acetone and acetoacetate. D-3-hydroxybutyrate is the most abundant ketone produced during ketosis the measurement of this analyte is more sensitive and specific.

The Randox D-3-Hydroxybutyrate (Ranbut) assay provides a convenient method for the accurate detection of ketones from serum. Liquid ready for convenience D-3-Hydoxybutyrate, has exceptional correlation coefficient of r=0.9954 and an excellent precision of <3.5% CV.

Available for use on a wide range of clinical chemistry analysers, the test is easily automated delivering quantitative results and allowing levels to be monitored for signs of improvement. This assay is suitable for use in open channels, it can be used with a wide range of clinical chemistry analysers.

Calibrator and controls are also available from Randox Laboratories, delivering the complete testing package to market in the fight against global pandemic levels of Diabetes Ketones circulating in the Blood Type 2 diabetes mellitus (T2DM).

Randox Diabetes Panel

Randox is committed to advancing diabetes testing capabilities, to help combat the 'Silent Diabetes Pandemic'. The Randox Diabetes testing panel consists of ten assays including niche and superior performance assays that are compatible with a wide range of clinical chemistry analysers. Randox offer the full spectrum of diagnosis & monitoring diagnostic solutions and associated diabetes complications including; ketoacidosis, renal dysfunction, and metabolic status. Contact Randox for more information, we look forward to hearing from you.

References

1. World Health Organization (WHO). Diabetes. https://www.who.int/news-room/fact-sheets/detail/diabetes (accessed 13 April 2021)
2. Misra S, Oliver NS. Diabetic ketoacidosis in adults. British Medical Journal (BMJ) 2015; 351 https://www.bmj.com/content/351/(accessed 29 April 2021).
3. Hecht M. Ketosis vs. Ketoacidosis: What You Should Know. https://www.healthline.com/health/ketosis-vs-ketoacidosis (accessed 20 April 2021).
4. Mayo Clinic. Diabetic ketoacidosis. https://www.mayoclinic.org/diseases-conditions/diabetic-ketoacidosis/symptoms-causes/syc-20371551 (accessed April 2021).
5. Brooke J, Stiell M, Ojo O; Evaluation of the Accuracy of Capillary Hydroxybutyrate Measurement Compared with Other Measurements in the Diagnosis of Diabetic Ketoacidosis: A Systematic Review. Int J Environ Res Public Health. 2016 Aug 2313 (9)
6. Type 1 diabetes in adults: diagnosis and management; NICE Guidelines https://www.nice.org.uk/guidance/ng17 (accessed April 2021).
7. Klocker AA, Phelan H, Twigg SM, Craig ME. Blood beta-hydroxybutyrate vs urine acetoacetate testing for the prevention and management of ketoacidosis in type 1 diabetes: a systematic review. Diabetic Medicine 2013; 30 (7): 818-824.
8. Palermo N, Sadhu A, McDonnell M. Diabetic Ketoacidosis in COVID-19: Unique Concerns and Considerations. The Journal of Clinical Endocrinology & Metabolism 2020;
9. Li J, Wang X, Chen J, Zuo X, Zhang H, Deng A. COVID-19 infection may cause ketosis and ketoacidosis. Diabetes Obesity Metabolism 2020; 105 (8): 2819-28

RANDOX