The lipaemic index: clinical observations

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ABSTRACT

The lipaemic index can be used to assess whether or not blood samples are suitable for laboratory analysis. However, little is known about which patients have a raised lipaemic index. In this article we study patient demographics and serum lipid concentrations in samples showing a raised lipaemic index. Of the 4271 patient samples measured in the month of July 2014, a total of 310 had a lipaemic index ≥0.4. Blood samples showing a raised lipaemic index were studied in a retrospective patient case review of laboratory results. Overall, 7.3% of all samples measured had a raised lipaemic index ≥0.4. This study found that males were more likely to have a high lipaemic index (56%) and neonates were the group most frequently producing lipaemic samples (30.6%). The correlation between the lipaemic index and the triglyceride concentration showed an r^2 value of only 0.37 (r=0.61), and the correlation between cholesterol and lipaemic index showed an r^2 value of 0.16 (r=–0.41). Male and neonatal samples were most likely to show a raised lipaemic index. There was a positive correlation between sample triglyceride and lipaemic index and an inverse correlation with cholesterol concentration and the lipaemic index, although this did not account for all the variance. Thus, other factors may also be important in the expression of the lipaemic index.

KEY WORDS: Amylases.
Hyperlipidemias.
Hypertriglyceridemia.
Lipemic index.

Introduction

Lipaemia in a patient blood sample is caused by an excess of lipoprotein particles, in particular chylomicrons and very low density lipoproteins (VLDL), which cause turbidity and can interfere with numerous biochemical methodologies.[1] This can have serious consequences for patients who may receive an incorrect diagnosis and unnecessary treatment.[2,3] Samples may be lipaemic for a number of reasons (e.g., taken soon after a high-fat meal or due to a hyperlipidaemia of either primary of secondary cause).[4]

Lipaemia is capable of interfering in analytical methodologies such as the use of ion-specific electrodes (ISEs), spectrophotometry, immunoassay and electrophoresis. Spectrophotometric analyses are most commonly affected due to the absorbance of light by lipoprotein particles. The amount of light absorbed decreases as the wavelength increases and therefore analyses that use low wavelengths are most affected by lipaemia. Spectrophotometric methods using a wavelength of 340 nm for measurement are therefore prone to this type of interference.

One of the most commonly affected analyses is that of sodium measurement by indirect ISE, in which lipaemia causes a volume displacement in the reaction cell, resulting in an aberrantly low sodium concentration result.[5] This interference can be overcome by the use of direct ISE, which does not dilute the sample and therefore does not suffer from the interference seen in indirect sodium measurement.

Amylase analysis can also be negatively affected by the presence of lipaemia, which can cause a diagnosis of acute pancreatitis to be missed. This is especially troubling as high serum triglyceride is a risk factor for acute pancreatitis, and therefore samples from acute pancreatitis patients may be lipaemic and mask raised amylase activity.[6]

It is important that the biochemistry laboratory recognises lipaemia in samples and is aware of the analytical methods that may be affected. It may be beneficial to the patient to add on lipid measurements to such samples, and measure analytes such as sodium by methods not affected by lipaemia (i.e., direct ISEs).

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The present study investigates lipaemic samples passing through the biochemistry laboratory during the month of July 2014. The aim of the investigation is to determine which patient groups most commonly showed a high lipaemic index (L-index) and whether or not this correlated with serum lipid results.

Materials and methods

Of the 4271 patient samples measured in the month of July 2014, a total of 310 had a lipaemic index ≥0.4. All samples with a lipaemic index ≥0.4 as measured on an Abbott Architect analyser were examined for patient demographics including clinical condition, lipid concentrations, sodium concentration and amylase activity and comments added automatically by the laboratory information management system (LIMS) or manually by laboratory staff. Samples were analysed on an Abbott Architect analyser with assay coefficient of variation (CV%) <5%.

From the laboratory clinical records, the most common causes of increased lipaemic index were noted. Laboratory data were collected on a Telepath pathology patient management system. The clinical audit office at the hospital approved this project. Ethical committee approval was not required as this was a retrospective observational study with no clinical intervention, although we did have agreement to do the study from the hospital audit department. Data were analysed using Microsoft Excel statistics software, and statistical significance was set at $P<0.05$.

Results

Of all samples measured in this study, 310 (7.3%) had a lipaemic index ≥0.4, which is higher than the average estimates in previous studies (0.5–2.5).[3,7] The patients involved were grouped by age and gender to determine the common characteristics of patients producing lipaemic samples. The results can be seen in Table 1 and indicate that males are more likely to have a high lipaemic index (56%) and that neonates or children aged less than one year are the groups most frequently producing such samples (30.6%). Adolescents and the elderly (≥75 years) were least likely to have a high lipaemic index (7.1% and 4.8%, respectively).

The locations of patients who had a raised lipaemic index were investigated and the results can be seen in Table 2. Data show that the majority of patients producing high lipaemic index samples presented to accident and emergency (A&E; 23.5%) or were hospital in-patients (20.6%). There was also a high number of neonatal intensive care unit (ITU) patients with a high lipaemic index (18.1%).

The clinical details on request forms or in the laboratory computer system were noted for each lipaemic sample. Of those patients with clinical details, the pathologies included those shown in Table 3. Where data were available, the correlation between the lipaemic index and serum lipids was studied; serum triglyceride concentration showed an r^2 value of 0.37 ($r=0.61$, $P=<0.0001$) and the correlation between serum cholesterol concentration and lipaemic index showed an r^2 value of 0.16 ($r=-0.41$, $P=0.0018$) (Fig. 1).

Discussion

This laboratory review investigated the frequency of lipaemia, as judged by a lipaemic index ≥0.4, in samples received over a period of a month in a large district general hospital. This study has investigated the characteristics of patients presenting with lipaemic samples, and some interesting findings have resulted from this work. There appears to be a higher frequency of lipaemic samples coming through our laboratory (7.3%) that is higher than reported elsewhere.[3,4]

Lipaemic samples were regarded as those having a lipaemic index ≥0.4 on an Abbott Architect analyser. When looking at patient demographics it has been shown that males are more likely to have a raised lipaemic index and that neonates form the largest group of patients producing lipaemic blood samples. The cause for these patient populations to have different lipaemic indices may be complex. It was noted that some of the neonatal samples with a raised lipaemic index were receiving total parenteral nutrition (TPN), which may be the cause of their hyperlipidaemia. Interestingly, some of these neonates tended not to have significantly raised serum triglycerides or cholesterol (Table 4), so the cause of the raised lipaemic index must be due to other causes. Previous studies have identified several causes of an unexpectedly raised lipaemic index, which have included abnormal proteins such as M proteins in myeloma patients and the presence of contrast dyes used in clinical investigations.[8,9]

The majority of the lipaemic samples came from A&E, in-patients or neonatal ITU. From the information available, the most common clinical details given on the pathology request forms were diabetes mellitus, health review, hypertension, abdominal pain, and gout. Diabetes mellitus is to be

Table 1. Patients involved were grouped by age and gender to determine the common characteristics of those producing lipaemic samples.

Gender	Number (%)
Male	173 (56)
Female	137 (44)
Age (years)	
<1	95 (30.6)
1–15	22 (7.1)
16–39	59 (19)
40–59	59 (19)
60–74	60 (19.4)
≥75	15 (4.8)

Table 2. Clinical locations of patients with a lipaemic index ≥0.4.

Location	Number (%)
GP	34 (10.9)
A&E	73 (23.5)
Paediatric A&E	22 (7.1)
In-patient	64 (20.6)
Out-patient	33 (10.6)
ITU	22 (7.1)
Neonatal ITU	56 (18.1)
Not stated	6 (2.3)

Table 3. Clinical conditions present in patients with lipaemic blood samples.

Clinical detail/reason for blood sampling	Frequency (%)
Diabetes mellitus	6
Health review	5
Hypertension	4
Abdominal pain	3
Gout	3
Tired	2
Pregnancy	2

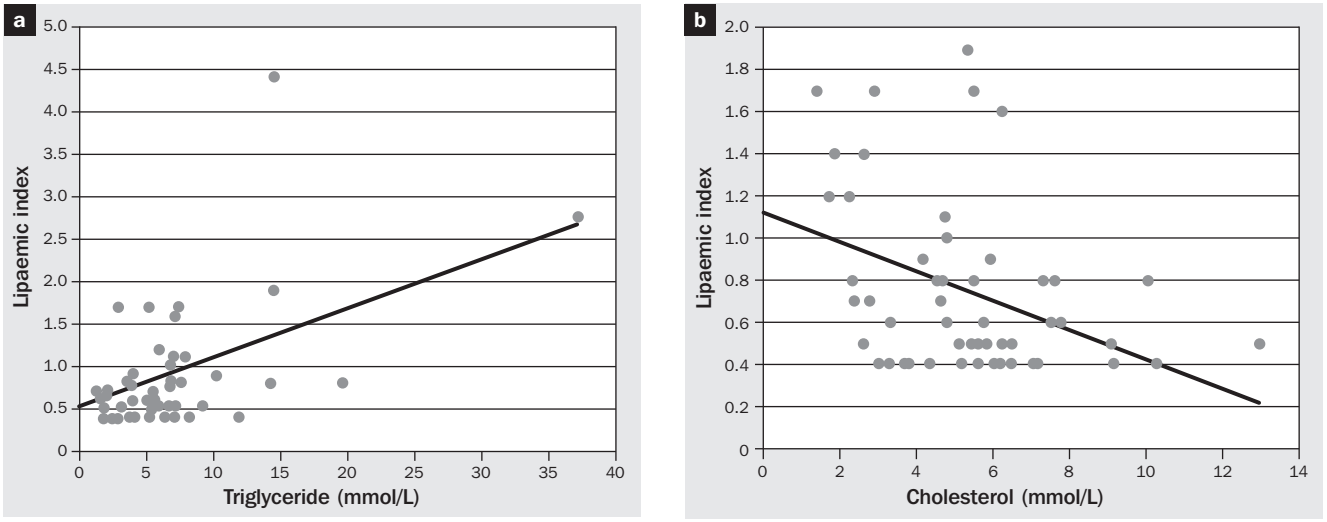

Fig. 1. Relationship between lipaemic index and **a)** serum triglyceride; **b)** cholesterol concentration.

expected as a common cause of secondary hyperlipidaemia and hypertension is associated with hyperlipidaemia.[10] Abdominal pain could indicate acute pancreatitis, for which hypertriglyceridaemia is a risk factor,[6] and hyperlipidaemia is a common finding in gout.[11] Interestingly, neonatal ITU samples seem particularly prone to this, although we do not have adequate explanation as to why this may be the case; it is possible that the use of TPN containing lipid emulsion could be contributory in some cases.[3,12]

The laboratory protocol following this study now states that all samples with a lipaemic index ≥0.4 should have a full lipid profile automatically added by the analyser; lipaemic index calculations on the Abbott Architect are based on turbidimetry measurements using dilution of the sample in saline or buffer and then measurement of spectra over a wide range of wavelengths. Lipaemic samples usually absorb light between 300 nm and 700 nm. The Abbott Architect system uses various wavelengths (i.e., 510/524; 572/604; 628/660 and 524/804 nm).[3] However, the automatic detection of lipaemia using the lipaemic index lacks standardisation among different analysers.[3]

The relationship between triglyceride concentration and lipaemic index has been investigated in several studies previously and shown to be highly variable. The source of triglyceride (i.e., endogenous lipoproteins or intravenous lipid emulsions) has been suggested as a cause of differing turbidity.[3] The relationship between serum triglyceride and cholesterol with the lipaemic index is shown in Figure 1. The correlation between the lipaemic index and the triglyceride concentration and cholesterol concentration showed r^2 values of 0.37 and 0.168, respectively. This supports and adds to the thorough paper of Twomey and colleagues,[7] although they did not report cholesterol concentration and they were looking also at visual appraisal of lipaemic samples.

Visual appraisal of the sample is not sufficient to detect lipaemia, not least because of operator variation.[1,7,13–15] Various methods to 'clear' the lipaemic samples, such as ultracentrifugation or the use of a detergent 'clearing' agent (e.g., LipoClear or n-hexane), may result in certain assay problems, although ultracentrifugation (approximately 10,000 *xg*) is probably preferable, if available,[14–18] although this, too, may be associated with assay difficulties such as those associated with hydrophobic substances, which may become distributed in the lipid layer and falsely decreased in the infranatant.

Clinicians should also be made aware of the limitations of biochemical analysis on highly lipaemic samples by the addition of interpretative comments on the final biochemical report, should the sample be found to show hypertriglyceridaemia. We use the following report comments in our laboratory for lipaemic samples: i) 'Serum triglyceride >10 mmol/L, exclude secondary causes and is there a family history of hyperlipidaemia?' ii) 'Severe hypertriglyceridaemia is a risk factor for acute pancreatitis, contact lipid specialist (plus his contact details given); iii) 'Serum triglyceride >20 mmol/L, sample not suitable for analysis, if suspicion of pancreatitis, please send urine amylase; iv) Sodium measured by method not affected by high lipids (in the case of the latter, a direct ion-selective electrode was used for serum sodium concentration determination).

In conclusion, laboratories should be aware of the problems that lipaemic samples can cause to certain assays, and they need to have policies in place to address this and

Table 4. Lipid profile details of four children aged less than two months with lipaemic index >1.0.

Lipaemic index	Serum triglyceride (mmol/L)	Serum cholesterol (mmol/L)	Clinical detail
1.4	1.7	2.6	Pre-term
1.4	0.93	1.86	Unwell ?cause
1.2	2.02	2.2	High lactate, sepsis
1.2	2.63	1.7	Total parenteral nutrition

appropriate clinical interpretative comments to instruct clinicians. The lipaemic index shows correlation with sample lipid concentrations, as shown in this study, although other factors are also implicated. We also report on which clinical samples and patient groups may be more susceptible to a raised lipaemic index; interestingly, samples from children less than a year old, including neonatal ITU samples, seem particularly prone to this, although we do not have adequate explanation as to why this may be the case; it is possible that the use of TPN containing lipid emulsion could be contributory in some cases. □

Important points

1 The lipaemic index shows positive correlation with serum triglyceride and inverse correlation with cholesterol concentration, but serum lipids do not account totally for the variability and thus other factors are important.
2 A raised lipaemic index was found to be relatively common in samples from males and neonatal ITU and children <1 year of age.
3 Overall, 7.3% of all biochemistry laboratory blood samples had a raised lipaemic index ≥0.4.

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