1. Introduction

Retinol-binding protein (RBP) has been reported to identify insulin resistance and associated cardiovascular risk factors. In one study circulating levels correlated positively and significantly with body mass index (BMI) and fasting insulin in subjects with a variety of clinical presentations, while subjects with diabetes had RBP levels two to three times above lean controls without diabetes [1]. Another study reported a less marked elevation of circulating RBP in patients with diabetes, despite an association with insulin resistance [2], although shortcomings in methodology may have complicated the measurement of RBP in these groups [3]. Promintzer et al. found no association between circulating RBP and insulin resistance [4], and circulating levels have also been reported to be no different between normal, overweight and obese subjects [5]. We recently reported the plasma variation of RBP in a small group of 10 matched normal and insulin-resistant subjects, and suggested that the link between plasma RBP and insulin resistance was tenuous [6]. We therefore extended this study, using the largest and most comprehensive cohort to date, and confirm that RBP is unlikely to be a useful marker of insulin resistance.
ELISA using polyclonal and monoclonal antibodies [6], native insulin using an immunoassay analyser (IMX, Abbott Laboratories, Illinois, U.S.A.) and glucose using the Aeroset analyser (Abbott). Insulin resistance (HOMA-R) was calculated using an established method [7]. Analysis of data was carried out using Sigmastat 3.1 software. Comparison between groups was by the un-paired \( t \)-test or the Mann–Whitney \( U \)-test when data was not adequately normal. The association between RBP and other variables were calculated using Spearman’s Rank Correlation coefficient.

3. Results

Plasma RBP levels are shown plotted against BMI and insulin resistance (HOMA-R) (Fig. 1A and B) with different symbols signifying the two groups. Whether the data was combined, or analysed group-wise, no association between RBP levels and either BMI or insulin resistance was observed (Pearson product moment correlation for combined data \( r = 0.003 \) and 0.071 respectively). Furthermore, there was no relationship between RBP and fasting insulin, fasting glucose, % body fat or waist circumference when the data was combined or analysed group-wise (combined data \( r = 0.050, 0.043, 0.082, 0.043 \) and \(-0.031\), respectively). The levels of RBP, fasting insulin, fasting glucose, HOMA-R, BMI, % body fat and waist circumference in the two groups are shown in Table 1. RBP levels were not significantly different between the two groups and similar to values reported previously [6], whereas, HOMA-R and fasting glucose, and to a lesser extent waist circumference, were significantly greater in subjects with diabetes.

4. Discussion

The results support the notion that plasma RBP is not a useful marker of insulin resistance. They extend our previous small study considerably where subjects with the metabolic syndrome had mean circulating RBP levels that were not significantly different than lean controls. Importantly the reference change values and the index of individuality of plasma RBP are five times greater in metabolic subjects than lean controls [6], implying that the metabolic syndrome cohort is less stable with respect to RBP. This is likely to further confound any relationship between circulating RBP and insulin resistance, as is the degree of renal impairment. Glomerular dysfunction in chronic renal failure can lead to increased circulating RBP levels [8,9] and tubular dysfunction can lead to decreased resorption and elevated RBP excretion, thereby compromising serum levels [9]. In a recent study Raila et al., using matched plasma and urine samples [10], reported that plasma RBP levels in type 2 diabetics were affected by incipient nephropathy. This further tempers consideration of RBP as a marker of insulin resistance.

![Fig. 1](image_url) - Correlation of plasma retinol-binding protein levels with (A) body mass index and (B) insulin resistance (HOMA-R). Groups were: (○) subjects with diabetes mellitus (n = 63) and (△) subjects without diabetes mellitus and with varying degrees of insulin resistance (n = 222).

### Table 1 - Data comparison between subjects with diabetes and those without diabetes and varying degrees of insulin resistance (mean ± S.D.)

<table>
<thead>
<tr>
<th></th>
<th>Subjects with diabetes (n = 63)</th>
<th>Subjects without diabetes (n = 222)</th>
<th>Mann–Whitney U-test or un-paired t-test (when data not adequately normal)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Waist circumference (cm)</td>
<td>107.5 ± 14.8</td>
<td>103.6 ± 65</td>
<td>( p = 0.008 )</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>31.7 ± 6.5</td>
<td>30.4 ± 8.7</td>
<td>( p = 0.089 )</td>
</tr>
<tr>
<td>Fasting insulin (pmol/L)</td>
<td>73.7 ± 58.2</td>
<td>71.1 ± 71.1</td>
<td>( p = 0.323 )</td>
</tr>
<tr>
<td>Fasting glucose (mmol/L)</td>
<td>8.1 ± 3.3</td>
<td>5.2 ± 0.9</td>
<td>( p &lt; 0.001 )</td>
</tr>
<tr>
<td>HOMA-R</td>
<td>4.6 ± 4.9</td>
<td>2.9 ± 3.2</td>
<td>( p &lt; 0.001 )</td>
</tr>
<tr>
<td>% body fat</td>
<td>36.8 ± 8.9</td>
<td>29.3 ± 10.8</td>
<td>( p = 0.123 )</td>
</tr>
<tr>
<td>RBP (mg/L)</td>
<td>39.9 ± 13.8</td>
<td>37.2 ± 12.5</td>
<td>( p = 0.125 )</td>
</tr>
</tbody>
</table>
Circulating RBP is often referred to as RBP4 [1–5,10]. However, its analysis usually employs antibodies generated against urinary RBP, which often serves as the standard in immunoassays. Although RBP can be adipocyte-derived it cannot be easily distinguished from the predominant hepatic synthesis of RBP or from minor immunoreactive post-translationally processed forms of RBP which also circulate [8], adding further to the confusion. Accordingly we suggest that the term RBP reflects what is actually measured in the circulation.

In this largest study cohort to date the mean plasma level of RBP in the diabetes mellitus group was only marginally higher than the mean level in subjects without diabetes and there was no significant association between plasma RBP and either BMI, or fasting insulin. We therefore consider the link between plasma RBP and insulin resistance is tenuous and that the measurement of circulating RBP is not a useful biochemical marker of insulin resistance.

Conflict of interest

The authors state that they have no conflict of interest.

REFERENCES


