

# A Multi-Marker Risk Score Predicts Insulin Sensitivity and Beta Cell Function-Driven Type 2 Diabetes From a Fasting Blood Sample

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## Aim

The goal of this study was to evaluate the degree to which models using biomarkers measured in a single plasma sample from fasting patients could substitute for the disposition index, and to understand how these fasting biomarker models relate to insulin sensitivity and secretion.

## Introduction

Insulin resistance and a compensatory increase in insulin secretion is the basis of glucose homeostasis. Type 2 diabetes results when pancreatic  $\beta$ -cells lose the ability to secrete sufficient insulin to compensate for increasing insulin resistance. Both insulin resistance and insulin secretion can be calculated from a frequently sampled intravenous glucose tolerance test (FSIGTT) using the minimal model (1). It has been shown that an increase in insulin resistance (or decrease in insulin sensitivity ( $S_I$ )) and/or a decrease in insulin secretion (as measured by acute insulin response (AIR)) occurs in advance of the development of diabetes. The interaction of  $S_I$  and AIR, called the disposition index, has previously been reported to be the best predictor of the development of incident diabetes (2).

In this study, we had access to baseline plasma specimens from subjects in the Insulin Resistance Atherosclerosis Study (IRAS) without diabetes at the study initiation who had complete minimal model measurements and who participated in the 5 year follow-up examination. These samples provided a unique opportunity to evaluate three biomarker models based on measurements in fasting samples to determine the extent to which they could capture the biological information provided by the minimal model. The models were: homeostatic model assessment, HOMA-IR and HOMA-%B, and Diabetes Risk Score (DRS). HOMA-IR and HOMA-%B, based on fasting measures glucose and insulin, are often used as surrogates for the biology of insulin sensitivity and

$\beta$ -cell function, respectively. DRS is a multi-marker model for assessment of diabetes risk based on measure of seven biomarkers in a fasting sample (3).

## Methods

**Subjects.** The selection of subjects in IRAS cohort has been previously described (4). The study included non-Hispanic whites, African Americans, and Hispanics. Participants were recruited to represent a range of glucose tolerance and ages spanning 40 to 69. A follow-up exam was conducted at an average of 5.3 yrs (4.6 to 6.6 years). For the current study, baseline plasma specimens were available from 695 subjects without diabetes at the study initiation who had complete minimal model measurements and who participated in the 5 year follow-up examination. During the follow-up period 117 subjects developed diabetes based on the 1999 WHO diagnostic criteria: fasting plasma glucose  $\geq 7.0$  mmol/l (126 mg/dl) or 2-h plasma glucose  $\geq 11.1$  mmol/l (200 mg/dl) (5). Baseline characteristics of the subjects are in shown in Table 1. There were significant differences between converters and non-converters in all of the characteristics presented except gender, race/ethnicity and diastolic blood pressure.

Insulin sensitivity, expressed as the  $S_I$ , was calculated using mathematical modeling methods (MINMOD version 3.0) (6). First-phase insulin secretion, expressed as the AIR, was defined as the mean increment in the plasma insulin concentration above basal at 2, 4 and 8 minutes after the administration of glucose.

HOMA-IR and HOMA-%B were calculated using the 1996 HOMA2 computer model, downloaded from [www.OCDem.ox.ac.uk](http://www.OCDem.ox.ac.uk).

In this study, the DRS was calculated in the IRAS cohort using the same model and coefficients that were determined for the PreDx<sup>®</sup> DRS in the Inter99 cohort (3), without refit.

Because HbA1c values at baseline were not available for the IRAS cohort, data from the 2001–2008 NHANES surveys (7) were used to estimate an appropriate mean HbA1c of 5.4% for use in the model.

## Results

### Discrimination Analysis

Receiver operator characteristic (ROC) curves for the HOMA measures, the DRS model, and the minimal model measures are shown in Figure 1.

DI was the best predictor of incident diabetes (AROC = 0.815). Of the three models based on fasting biomarkers, DRS was the best predictor of incident diabetes (AROC = 0.761). DRS was also superior to either  $S_1$  or AIR alone.

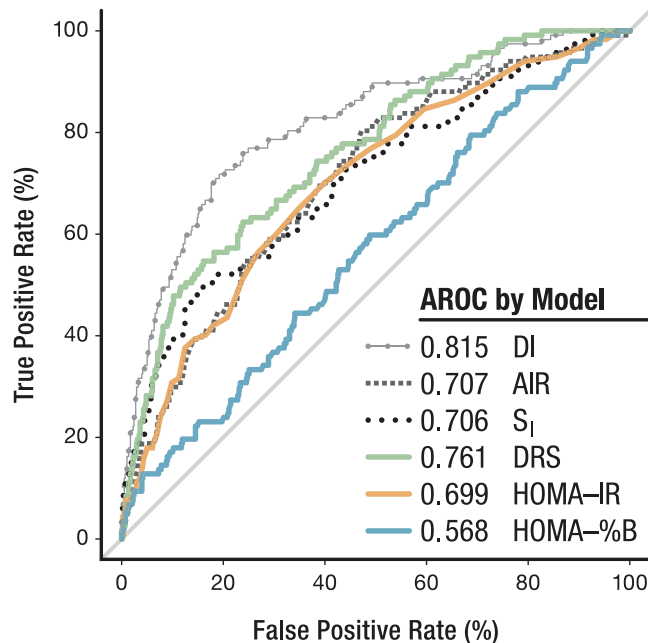
### Correlation with $S_1$ , AIR, DI

In Figure 2, HOMA-%B, HOMA-IR and DRS values are each plotted against AIR,  $S_1$  and DI. The correlation between each pair of measures was evaluated by Spearman's rank correlation coefficient. HOMA-%B is meant to act as a surrogate measure of beta-cell function or insulin secretion, and 17% of its rank variance may be explained by correlation with AIR, greater than either HOMA-IR (5%) or DRS (2%). The  $r_s^2$  value of HOMA-IR with  $S_1$  was 44%, consistent with its objective as a surrogate measure of insulin resistance, but 33% of the rank variance in DRS, and 22% in HOMA-%B, were also shared with  $S_1$ . DRS was the most strongly related to DI, with an  $r_s^2$  of 33%, compared to 12% for HOMA-IR and less than 1% for HOMA-%B.

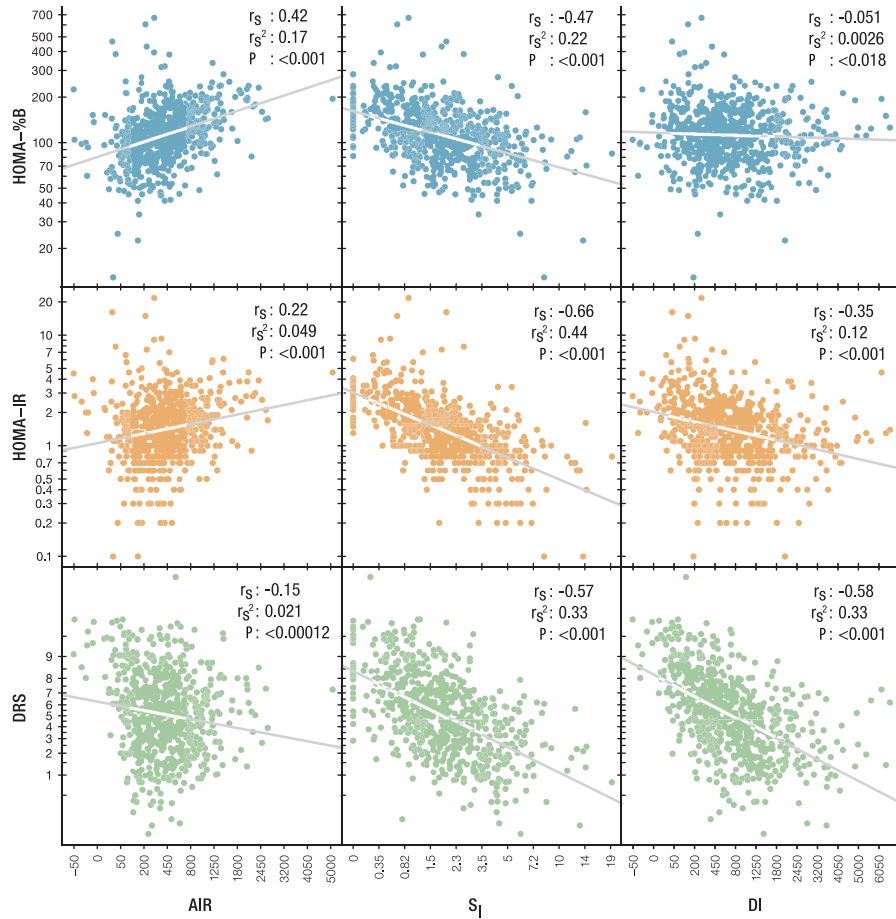
**Table 1.** Baseline characteristics of the study population by diabetes conversion status at the 5-year follow up visit. Data are n (% of stratum) or median (inter-quartile range). P-values were calculated by chi-squared (factors) or Wilcoxon rank sum test (continuous variables). NFG, normal fasting glucose; IFG, impaired fasting glucose; NGT, normal glucose tolerance; IGT, impaired glucose tolerance; HOMA, homeostatic model assessment.

Characteristic	Nonconverters	Converters	P-Value
<i>n</i>	578 (83.2%)	117 (16.8%)	
Sex			0.6
female	309 (82.4%)	66 (17.6%)	
male	269 (84.1%)	51 (15.9%)	
Race or Ethnicity			0.9
Non-Hispanic White	240 (82.8%)	50 (17.2%)	
Hispanic	201 (83.1%)	41 (16.9%)	
African-American	137 (84.0%)	26 (16.0%)	
Fasting glucose status			<0.001
NFG (<5.6 mmol/L)	452 (89.0%)	56 (11.0%)	
IFG (5.6 – 6.9 mmol/L)	126 (67.4%)	61 (32.6%)	
2-h glucose status			<0.001
NGT (<7.8 mmol/L)	425 (91.0%)	42 (9.0%)	
IGT (7.8 – 11.0 mmol/L)	153 (67.1%)	75 (32.9%)	
Age (years)	54.3 (47.6 – 62.3)	57.0 (49.7 – 63.9)	0.03
BMI (kg/m <sup>2</sup> )	27.0 (24.5 – 29.7)	29.4 (26.5 – 34.7)	<0.001
Waist circumference (cm)	89.3 (80.6 – 97.2)	94.0 (87.7 – 102.0)	<0.001
Systolic blood pressure (mm Hg)	118 (109 – 130)	125 (114 – 139)	<0.001
Diastolic blood pressure (mm Hg)	78 (71 – 84)	79 (72 – 85)	0.2
Fasting glucose (mmol/L)	5.1 (4.8 – 5.5)	5.6 (5.2 – 6.1)	<0.001
2-h glucose (mmol/L)	6.6 (5.3 – 7.9)	8.4 (7.3 – 10.1)	<0.001
Fasting insulin (pmol/L)	72 (48 – 102)	102 (78 – 150)	<0.001
Acute Insulin Response [pmol/(min.mL)]	428 (225 – 700)	162 (79 – 418)	<0.001
Insulin sensitivity [10 <sup>-4</sup> /(min.μU.mL)]	1.86 (1.05 – 3.22)	1.00 (0.48 – 1.70)	<0.001

**Figure 1.** Receiver operator characteristic (ROC) curves for the HOMA measures, the DRS model, and the minimal model measures. Areas under the ROC curve (AROC) are also tabulated as a measure of discrimination between converters and non-converters.



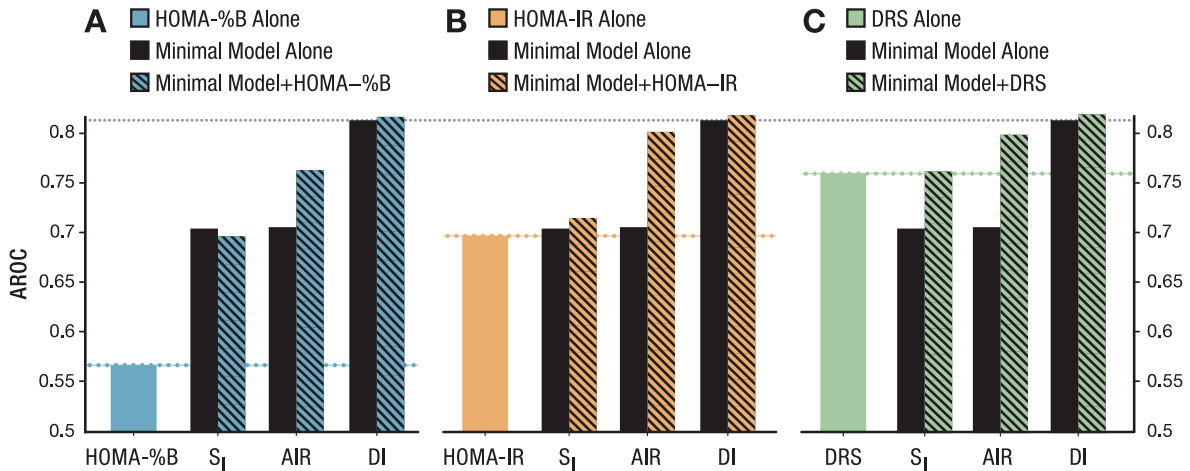
**Figure 2.** Spearman rank correlation coefficients ( $r_s$ ) were calculated between each of the minimal model measures, HOMA-IR and HOMA-%B, and the DRS. P-values were calculated assuming that  $r_s$  follows an approximate asymptotic t-distribution.



### Logistic Regression Analysis of Model Combinations

As a way of understanding how each of the fasting biomarker models relates to the underlying biology of AIR and  $S_1$ , we explored whether combining each of the fasting models with AIR or  $S_1$  in logistic regression models could match the predictive power of DI. Significant improvement in the AROC of the combination relative to the minimal model measures alone indicated that the measures were independent and complementary, while a lack of significant improvement indicated redundancy in terms of the physiology being measured.

**Figure 3.** The bootstrap cross-validated results of each single measure and each combination are presented in Figure 3.



**Table 2.** Table 2 shows the incremental change in the AROC for each combination versus each of its individual components. None of the combinations were significantly superior to DI alone (AROC=0.815; all P>0.2).

Model	AROC Alone	Incremental AROC From Combining Models					
		+ HOMA-%B	+ HOMA-IR	+ DRS	+ AIR	+ S <sub>1</sub>	+ DI
HOMA-%B	0.568	-	0.173 <sup>†</sup>	0.195 <sup>†</sup>	0.197 <sup>†</sup>	0.130 <sup>#</sup>	0.250 <sup>†</sup>
HOMA-IR	0.699	0.042 <sup>°</sup>	-	0.066 <sup>*</sup>	0.105 <sup>#</sup>	0.018	0.122 <sup>†</sup>
Diabetes Risk Score (DRS)	0.761	0.002	0.003	-	0.039 <sup>*</sup>	0.002	0.059 <sup>#</sup>
Acute Insulin Response (AIR)	0.707	0.058 <sup>*</sup>	0.096 <sup>#</sup>	0.093 <sup>#</sup>	-	0.105 <sup>#</sup>	0.104 <sup>#</sup>
Insulin Sensitivity (SI)	0.706	-0.008	0.011	0.058 <sup>*</sup>	0.106 <sup>#</sup>	-	0.106 <sup>#</sup>
Disposition Index (DI)	0.815	0.003	0.005	0.006	-0.004	-0.003	-

Significance of incremental improvement (p-values): 0 < † ≤ 0.001 < # ≤ 0.01 < \* ≤ 0.05 < ° ≤ 0.1 < ≤ 1

HOMA-%B was a poor predictor of future diabetes, with an AROC of 0.568 (Table 2). Although meant to be a surrogate of β-cell function, the combination of HOMA-%B with S<sub>1</sub> was not more predictive than S<sub>1</sub> alone.

HOMA-IR predicted incident diabetes with an AROC of 0.699 which was not significantly less than S<sub>1</sub> alone (Table 2). Combining S<sub>1</sub> with HOMA-IR did not significantly increase the AROC above either S<sub>1</sub> or HOMA-IR alone (Table 2, Figure 3), while in combination with AIR, it was as predictive as DI (Figures 3, Table 2). This indicates that HOMA-IR was a good surrogate for the biological role of insulin sensitivity in the progression towards diabetes.

As previously noted, DRS was significantly superior to either HOMA-%B (P<0.001) or HOMA-IR (P=0.01) alone. Adding S<sub>1</sub> to DRS did not significantly increase the AROC above the DRS alone (Table 2, Figure 3) while AIR combined with DRS (AROC=0.800) was significantly higher than DRS (P=0.04) or AIR (P=0.003) alone, and equivalent to DI alone (P=0.5).

## Conclusions

- In this study DI was the best predictor of incident diabetes.
  - However DI, based on measures obtained during an FSIGTT, is difficult to implement in routine clinical practice.
- HOMA-%B, HOMA-IR and DRS, based on measures in fasting blood samples, were assessed for their ability to predict incident diabetes compared to SI and AIR measured during an FSIGTT.
  - DRS was superior to HOMA-%B, HOMA-IR, SI or AIR alone
  - Combining SI with DRS did not significantly improve the prediction of diabetes over DRS alone
  - Combining DRS with AIR significantly increased the AROC to 0.800, equivalent to DI alone
- These results suggest DRS is an excellent surrogate for SI and appears to fully cover the biology of insulin sensitivity as it relates to assessment of diabetes risk.
- Additionally, the DRS partially covers the biology of b-cell function since it improves the prediction over SI alone.
- Overall, DRS was the best fasting surrogate for DI.

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