

# Glycated Albumin

PINFO 18/2015

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# Introduction to Glycated Albumin

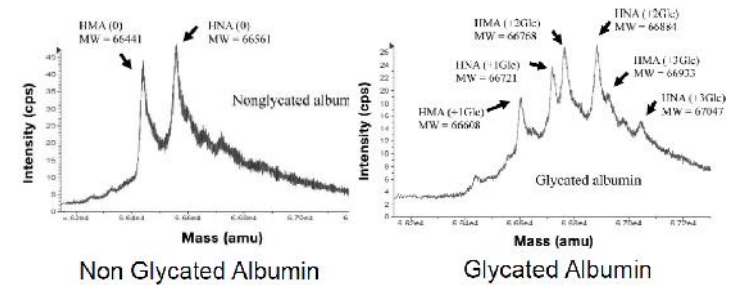
# Albumin: Protein synthesized in the liver

- Mol. Weight of 66 kDa
- Half Life: **17-23 days**
- Distribution in the body
  - ✓ 35-40% in plasma
  - ✓ 50-60% extravascular
- Concentration: 3.5-5.5 g/dL
- Functionality:
  - ✓ Maintenance of serous colloidal osmotic pressure
  - ✓ Carriage of poor solubility substances (i.e. fatty acid, bilirubin and drugs)
  - ✓ Anti-oxidant effect    Bourdon E et. al. The FASEB Journal 1999; 13: 233-244.

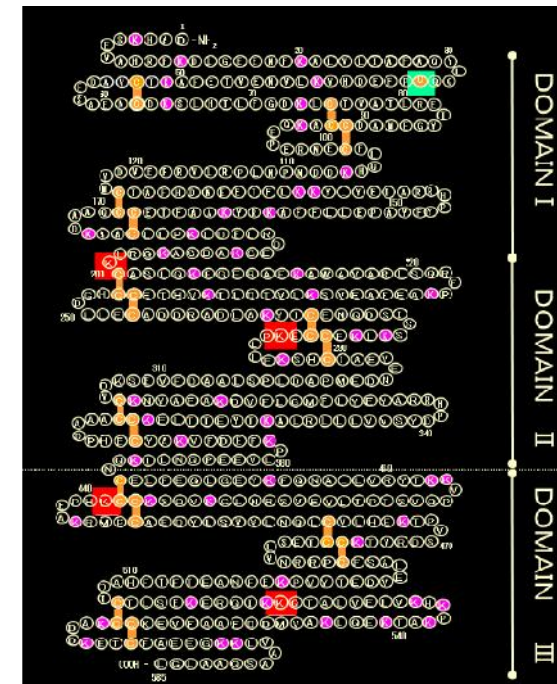
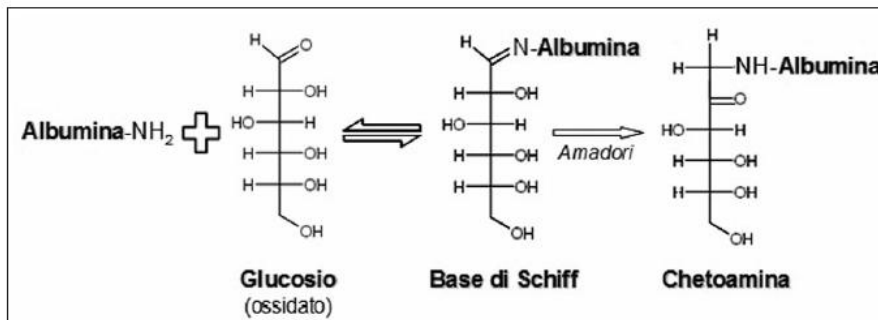
# Glycated Albumin (GA)

- Under hyperglycemia conditions all proteins are subject to a non enzymatic glycation process
- Albumin is one of the proteins more affected by glycation because of its high concentration and long half life
- **Glycated Albumin (GA)** represents 80% of the overall glycated proteins in human serum

Schleicher ED, Mayer R, Wagner EM et al (1988)  
 Is serum fructosamine assay specific for determination of glycated serumprotein? Clin Chem 34:320-323



Kohzuma et al. J Diabetes Sci Technol, 5(6) 2011



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# Albumin glycation effects

- Being Albumin distributed in the whole body, its Albumin glycation will **reflect glycation status of the whole body**
- Albumin glycation would **reduce drug binding properties and anti-oxidant properties**

Rondeau P, Bourdon E (2011) The glycation of albumin: Structural and functional impacts. *Biochimie* 93:645-658

Barnaby OS, Cerny RL, Clarke W et al (2011) Comparison of modification sites formed on human serum albumin at various stages of glycation. *Clin. Chim. Acta* 412:277-285

Bourdon E, Loreau N, Blache D (1999) Glucose and free radicals impair the antioxidant properties of serum albumin. *FASEB J* 13:233-244

Cohen MP, Shea E, Chen S et al (2003) Glycated albumin increases oxidative stress, activates NF-kB and extracellular signal-regulated kinase

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# Albumin glycation effects

- **High level of Glycated Albumin** may induce irreversible cellular damages partially responsible for **clinical complications of diabetes mellitus** (retinopathy, nephropathy, neuropathy, micro and macro vascular outcomes)

Cohen MP (2003) Intervention strategies to prevent pathogenetic effects of glycated albumin. Arch Biochem Biophys 419:25-30

Brownlee M (1995) The pathological implications of protein glycation. Clin Invest Med 18:275-281

Tang SC, Leung JC, Lai KN (2011) Diabetic tubulopathy: an emerging entity. Contrib Nephrol 170:124-134

Dobrian A, Simionescu M (1995) Irreversibly glycated albumin alters the physico-chemical characteristics of low density lipoproteins of normal and diabetic subjects. Biochim Biophys Acta 1270(1):26-35

Rubenstein DA, Yin W (2009) Glycated albumin modulates platelet susceptibility to flow induced activation and aggregation. Platelets 20:206-215

Unoki H, Yamagishi S (2008) Advanced glycation end products and insulin resistance. Curr Pharm Des 14:987-989

Shiraki T, Miura Y, Sawada T et al (2011) Glycated albumin suppresses glucose-induced insulin secretion by impairing glucose metabolism in rat pancreatic  $\beta$ -cells. Nutr Metab (Lond) 6:8-20

Turk Z, Ljubic S, Turk N et al (2001) Detection of autoantibodies against advanced glycation endproducts and AGE-immune complexes in serum of patients with diabetes mellitus. Clin Chim Acta 303:105-115

Natan et al. (7) Relationship of Glycated Albumin to Blood Glucose and HbA1c Values and to Retinopathy, Nephropathy, and Cardiovascular Outcomes in the DCCT/EDIC Study **Diabetes** Volume 63, January 2014

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# Glycated Albumin limitations

- In those clinical conditions which may influence Albumin levels (i.e. Thyroid disfunctions, Nephrotic syndrome, Cirrhosis, Non Alcoholic Steato-Hepatitis)

Okada T, Nakao T, Matsumoto H et al (2011) Influence of Proteinuria on Glycated Albumin Values in Diabetic Patients with Chronic Kidney Disease. Intern Med 50:23-29

Koga M, Murai J, Saito H et al (2010) Serum glycated albumin levels, but not glycated hemoglobin, is low in relation to glycemia in non-diabetic men with nonalcoholic fatty liver disease with high alanine aminotransferase levels. Clinical Biochemistry 43:1023-1025

- Other factors to consider: age, BMI, nutritional status, smoke, Hyperuricemia

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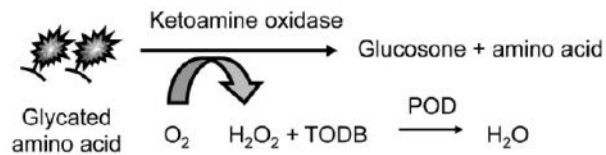
# Methods for the quantification of glycated serum proteins

Principio	Metodo	Pro / Contro
Colorimetrico	Acido tiobarbiturico Nitroblu di tetrazolio Fenilidrazina	Economico; poco accurato (fruttosamina) Come sopra
Enzimatico	Proteinasi K / Fruttosaminasi	Determinazione dei prodotti di Amadori; non sviluppato per uso clinico Semplice; misura le proteine glicate, ma con maggiore specificità dei metodi colorimetrici; unica azienda produttrice
Enzimatico	Proteasi Albumino-specifica / Chetoaminaossidasi / bromocresolporpora	Semplice e accurato; unica azienda produttrice, non approvato in Europa
Cromatografico	HPLC: Cromatografia di affinità (Boronato) preceduta da cromatografia a scambio ionico Furosina-HPLC	Preciso e accurato; costoso, possibile perdita di accuratezza in base all'efficienza separativa
Elettroforetico	Fenilboronato - PAGE	Poco accurato (fruttosamina), costoso
Immunometrico	ELBIA ( <i>enzyme-linked boronate immunoassay</i> ) ELISA anti-AGE Radio- <i>immunoassay</i>	Accurato; costoso Preciso; costoso, possibile inaccuratezza per l'alta specificità degli anticorpi monoclonali Come sopra
Spettrometria di massa	MALDI LC-MS	Come sopra Non sviluppato per uso clinico Non sviluppato per uso clinico
Spettroscopico Elettrochimico	Spettroscopia Raman FN6K <i>enzyme electrochemical sensing system</i>	Non sviluppato per uso clinico Non sviluppato per uso clinico

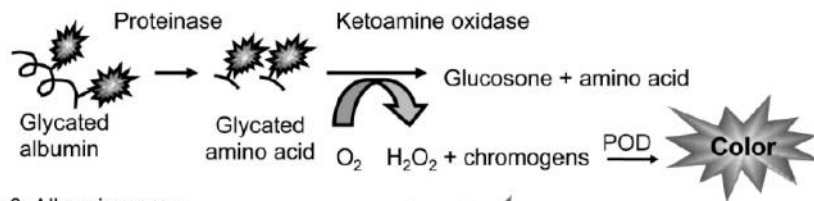
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# Method for the quantification of Glycated Albumin (GA)

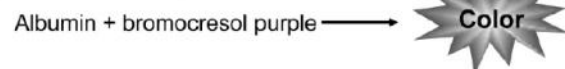
Step 1: Glycated amino acid elimination reaction



Step 2: Glycated albumin assay



Step 3: Albumin assay



Step 4: Calculation of percentage of glycated albumin in total albumin

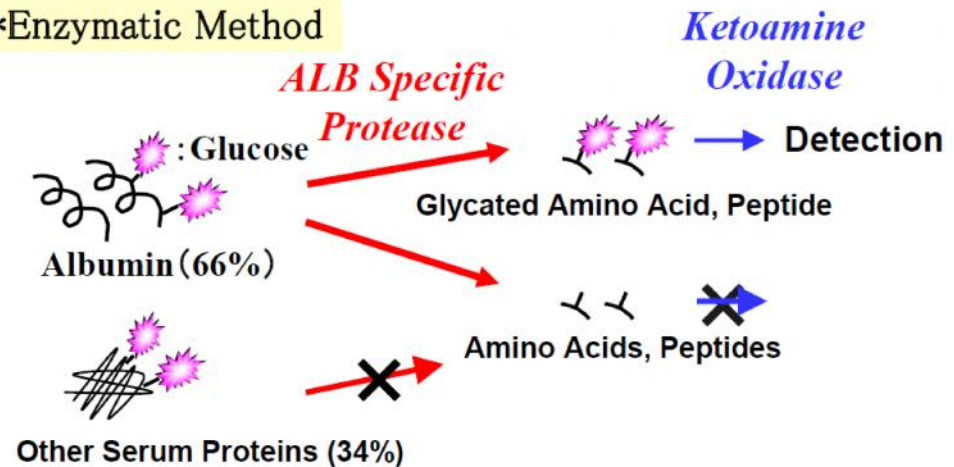
$$\text{GA value} = \frac{\text{GA concentration}}{\text{albumin concentration}} \times 100$$

Enzymatic method coupled to a colorimetric output by ketoamine oxidase and an albumin specific protease.

(developed by Asahi Kasei Pharma Corporation)

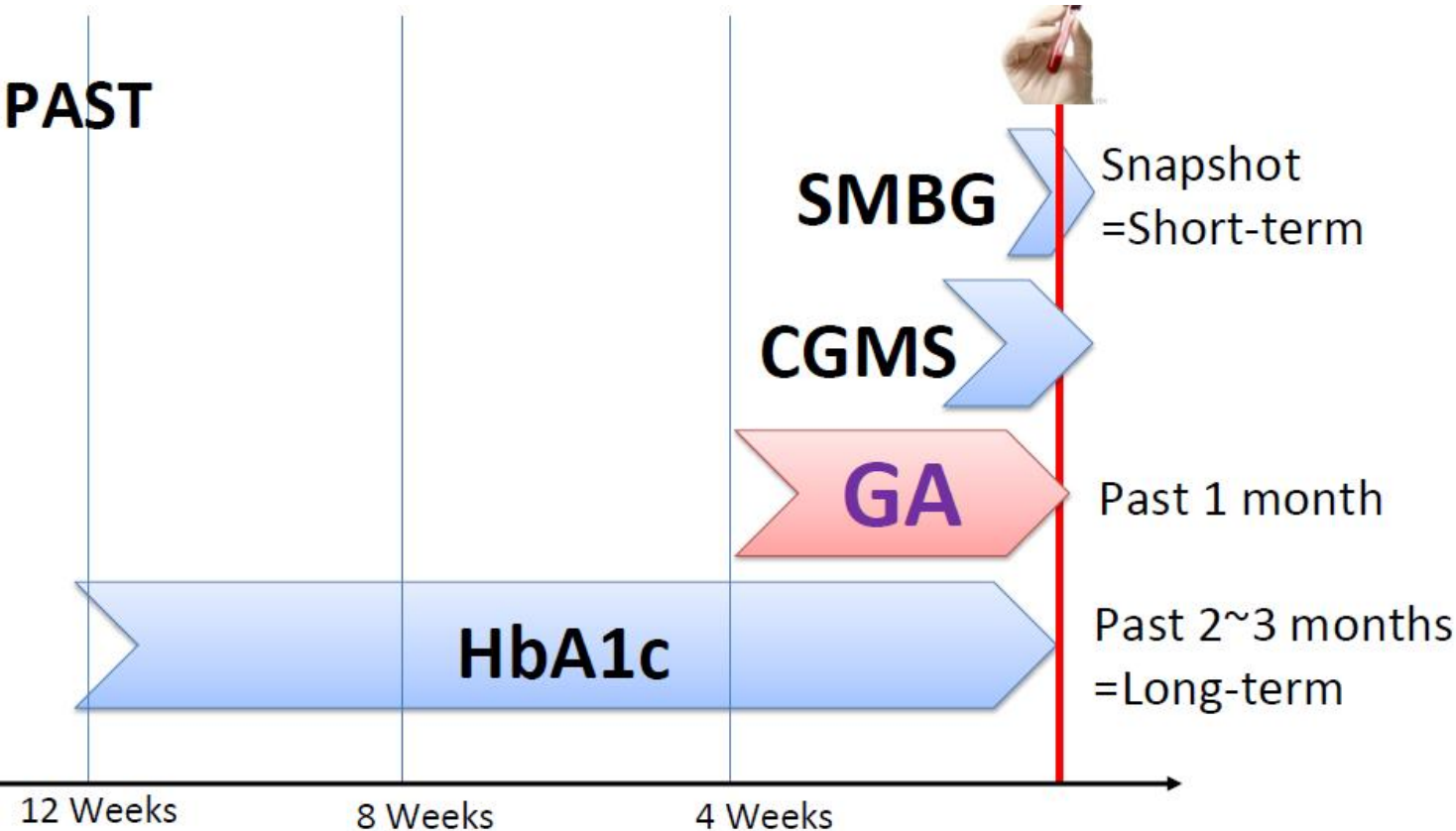
It is an **easy and reliable fully automated method** allowing accurate and precise determination of Glycated Albumin in a routine laboratory

## \*Enzymatic Method



# Glycated Albumin Clinical Value

# Hyperglycemia diagnosis and monitoring tools



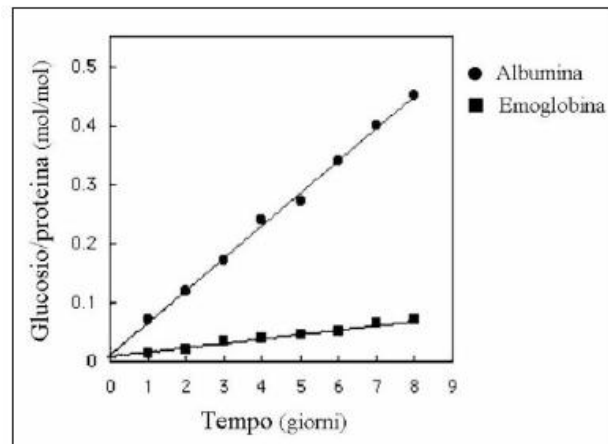
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# Glycated Albumin as a hyperglycemia diagnosis and monitoring tool

- **Short to intermediate integrated glycemic control** thanks to GA sensibility to short term glucose variations which might not be detected by fasting plasma glucose testing (single point) or disappear in the HbA1c testing (2-3 months average)

Takahashi S, Uchino H, Shimizu T (2007) Comparison of Glycated Albumin (GA) and Glycated Hemoglobin (HbA1c) in Type 2 diabetic patients: usefulness of GA for evaluation of short-term changes in glycemic control. *Endocrin Journal* 54(1):139-144

Won HK, Kim KJ, Lee BW et al (2012) Reduction in glycated albumin can predict change in HbA(1c): comparison of oral hypoglycaemic agent and insulin treatments. *Diabet Med* 29(1):74-79



**Albumin glycation rate is 10 times higher than Hb**

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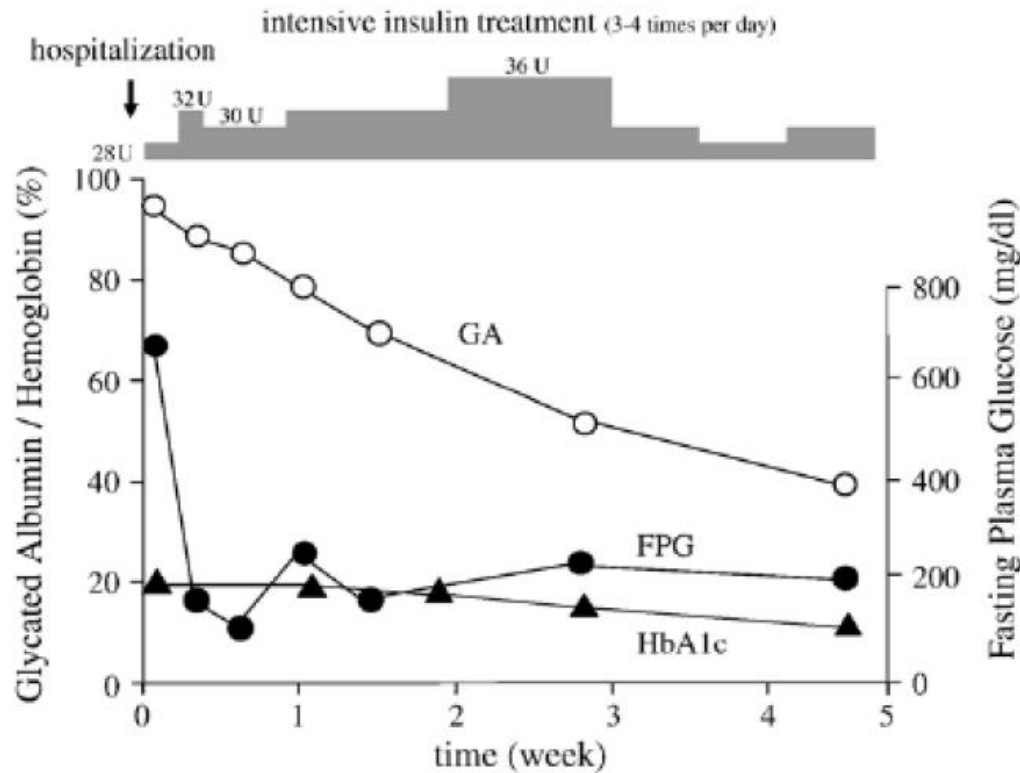
# Glycated Albumin as a hyperglycemia diagnosis and monitoring tool

- **Glycemic control in neonatal diabetes (NDM):** HbA(1c) is influenced by age-related changes in Fetal Hb and does not accurately reflect glycemic control

Glycated albumin but not HbA1c reflects glycaemic control in patients with neonatal diabetes mellitus. Suzuki S<sup>1</sup>, Koga M, Amamiya S, Nakao A, Wada K, Okuhara K, Hayano S, Sarhat AR, Takahashi H, Matsuo K, Tanahashi Y, Fujieda K. Diabetologia. 2011 Sep;54(9):2247-53. doi: 10.1007/s00125-011-2211-8. Epub 2011 Jun 5.

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# Glycated Albumin as a hyperglycemia diagnosis and monitoring tool



Patient ; female 58-y-old  
Presented at the  
emergency department

Symptom: fatigue,  
dyspepsia and faintness  
(Insulin therapy was  
stopped by herself)

BG; 671mg/dL, HbA1c  
19.5 %, GA 94.1%

Hospitalized and start  
intensive insulin therapy

Kisugi R, Kohzuma T, et al. Clin Chem Acta, 382;59-64, 2007 12

# Glycated Albumin in diabetic nephropathy

- GA is a glycemic control marker **more accurate than HbA1c in peritoneal dialysis and hemodialysis patients**
- Kidney deficiency is often associated to a reduction of erythrocytes  
half-life: **HbA1c may underestimate average glucose while GA doesn't**

Inaba M, Okuno S, Kumeda Y et al (2007) Glycated albumin is a better glycemic indicator than glycated hemoglobin values in hemodialysis patients with diabetes: effect of anemia and erythropoietin injection. J Am Soc Nephrol 18:896-903

Vos FE, Schollum JB, Coulter CV et al (2012) Assessment of Markers of Glycaemic Control in Diabetic Patients with Chronic Kidney Disease using Continuous Glucose Monitoring. Nephrology (Carlton) 17(2):182-188

Ma WY, Wu C, Pei D et al (2011) Glycated albumin is independently associated with estimated glomerular filtration rate in non diabetic patients with chronic kidney disease. Clinica Chimica Acta 412:583-586

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# Glycated Albumin in Gestational diabetes

- HbA1c decreases in the first quarter pregnancy, then it increases
- Non diabetic pregnant women show HbA1c levels lower than non pregnant women, due to glycaemia deficiency in the first quarter and iron deficiency in the second quarter
- Being GA independent from the above variations, it can be a **better tool for glycemic control of women with gestational diabetes**
- GA can also be useful to better monitor diabetes during pre-pregnancy

Phelps RL, Honig GR, Green D et al. (1983) Biphasic changes in haemoglobin A1c concentrations during normal human pregnancy. Am J Obstet Gynecol 147:651-653

# Glycated Albumin in Anemia and Hemoglobinopathy

- HbA1c underestimates average glicemia in case of anemia; therefore GA would be preferrable in terms of accuracy **for the monitoring of diabetic patients affected by anemia**
- This also applies to post hemorrhagies, hemolytic anemia, post transfusions e hemoglobinopathy

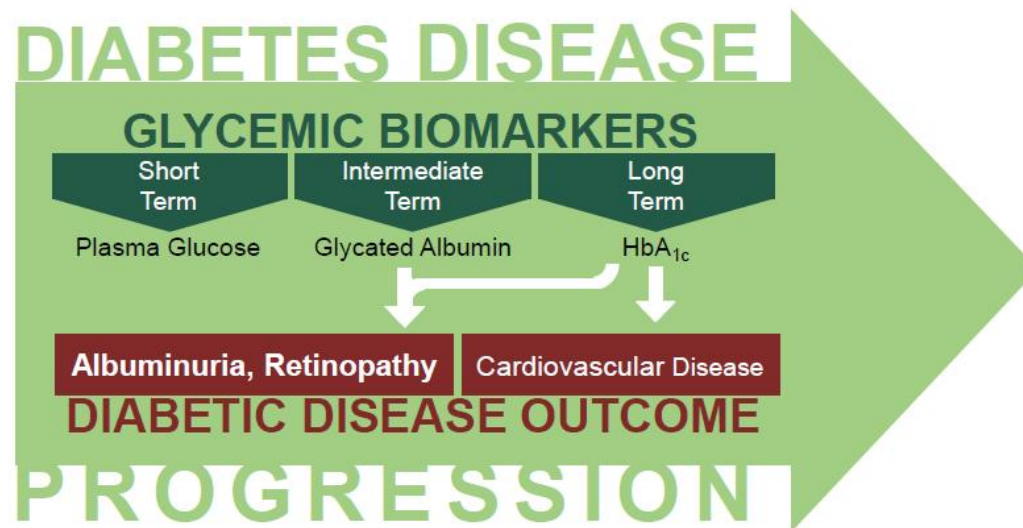
Kim S, Min WK, Chun S et al (2011) Glycated albumin may be a possible alternative to hemoglobin A1c in diabetic patients with anemia. Clin Chem Lab Med 49(10):1743-1747

Kim C, Bullard KM, Herman WH et al (2010) Association between iron deficiency and A1C Levels among adults without diabetes in the National Health and Nutrition Examination Survey, 1999-2006. Diabetes Care 33:780-785

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# Glycated Albumin and Diabetes Microvascular disease progression

**Figure 1** — Nathan et al. (7) provide compelling data to suggest the combination of glycated albumin and HbA<sub>1c</sub> strengthens the association with microvascular end points such as nephropathy (e.g., albuminuria) and retinopathy (e.g., nonproliferative retinopathy). The strength of this association was stronger than that for CVD and implies that a glycemic staging system may be especially useful for understanding diabetes microvascular disease progression such as kidney and eye disease.



Diabetes. 2014;63:46.

# Conclusions

- Diabetes is managed by clinicians with few tools coming from laboratory medicine
- Glycated Albumin is an analytically robust and reliable assay, easy to run on routine chemistry analyzers, with no need of sample pre-treatment
- Glycated Albumin is mid-term glycation indicator independent from Hb abnormalities, therefore can be placed side by side to HbA1c in all those clinical settings where HbA1c can be critical (renal failure, anemia, erythrocyte abnormalities, pregnancy)

# Conclusions

- In naïve diabetic patient diagnosis, Glycated Albumin could represent a further tool to assess glycation exposure in a shorter time frame
- Glycated Albumin can also find room in the pre-diabetic status assessment, where average glucose concentration is getting higher and higher, but HbA1c value is not yet affected

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