

CDT TEST IN SERUM BY UV-FAST-MONOREAGENT

Code Z68210

Code Z68215



CDT and its analysis
Analytical procedure
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IFU



September 2021

CARBOHYDRATE DEFICIENT TRANSFERRIN (CDT)

Exposure to large amount of alcohol in a chronic and heavy alcohol use can lead to alterations in the microheterogeneity of the iron transport glycoprotein transferrin.

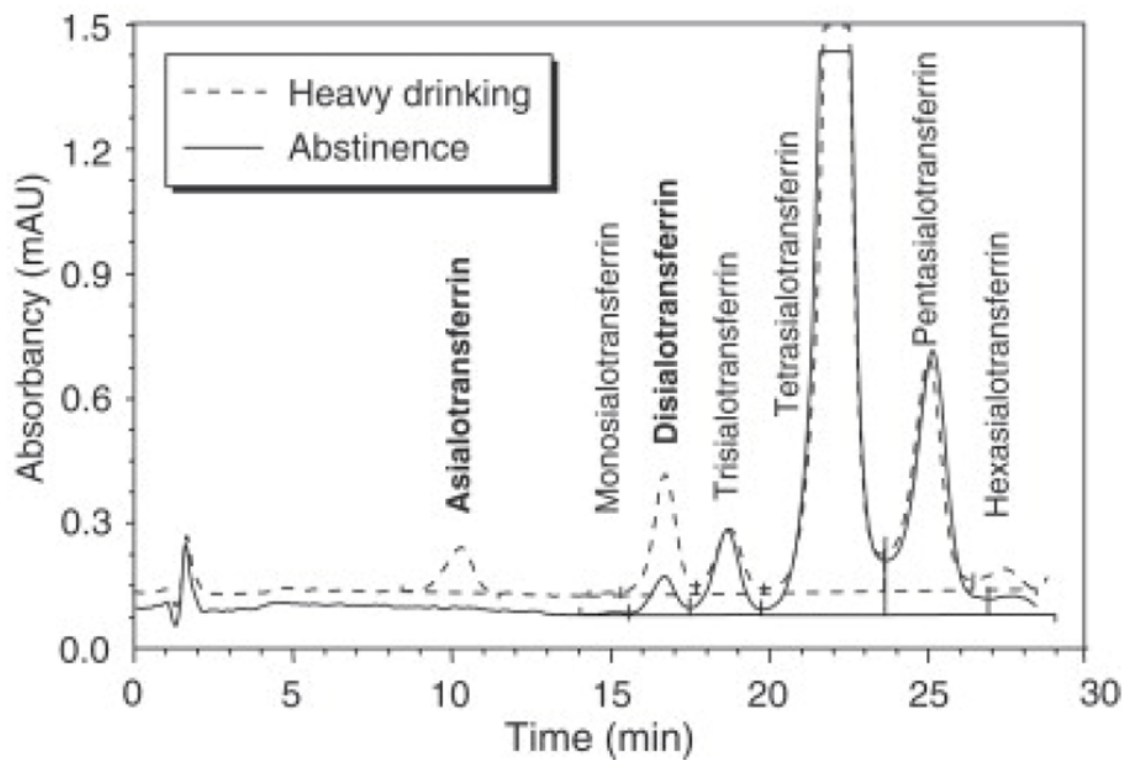
Structural modifications in transferrin have been observed, in cerebrospinal fluid (CSF), in serum sample from individuals with alcohol dependence and in patient suffering from congenital disorders of glycosylation (CDG) [1].

Liver function tests were also used as alcohol indicators. However, their low specificity for alcohol led to impossibility to discern alcohol dependent patients from ones suffering from non-alcohol-related hepatic pathologies.

Therefore, CDT test represents a valid method for the identification of chronic alcohol consumption and for screening and diagnosis of CDG.

Different transferrin glycoforms owe their name to the total number of terminal, negatively charged sialic acid residues. Tetrasialotransferrin is the most abundant among the different forms of transferrin in human serum (75–80% of total transferrin). Other common glycoforms are pentasialo- (~15%), trisialo- (~5%), hexasialo- (~2%), and disialotransferrin (~2%), whereas asialo-, monosialo-, heptasialo-, and octasialotransferrin usually are under detection limit. On the contrary, chronic alcohol abusers, typically have increased levels of disialotransferrin or asialotransferrin [1].

CARBOHYDRATE DEFICIENT TRANSFERRIN (CDT)



Taken from Dasgupta *et al.* Alcohol and its Biomarkers, SCIENCE DIRECT-2015.

Fig. 1. Comparison of levels of transferrin glycoform pattern in serum from an alcoholic patient after long period of heavy drinking and after abstinence from alcohol consumption.

The relative amounts of disialo- and asialotransferrin are increased in after heavy drinking. Taken from Dasgupta *et al.* Alcohol and its Biomarkers, SCIENCE DIRECT-2015.

ANALYSIS

Both asialo and disialotransferrin can be correlated to chronic alcohol consumption [2-3]. Nevertheless, the commission of IFCC has identified the disialotransferrin as target analyte for CDT. Although asialoform is more specific than disialotransferrin for alcohol abuse, it can only be identified by HPLC when levels of disialo increase. Therefore, disialotransferrin has the highest diagnostic relevance.

HOW TO EXPRESS THE RESULTS

There are different way to express CDT: the commission of IFCC suggests to calculate the percentage in relation to the total transferrin (%CDT), to cover false-positive results or false negative results linked to high or low value of total transferrin [4].

$$\%CDT = \frac{\text{Area}_{(\text{asialo} + \text{monosialo} + \text{disialotransferrin})}}{\text{Area}_{\text{total Trasferrin}}} \times 100$$

Total transferrin is calculated as Integration baseline: a-, mono-, di-, tri-, tetra-, pentasialotransferrina.

Associated with other tests such as transaminases, GGT and MCV, CDT can be a useful tool in identifying problem drinking i.e. chronic alcohol abuse or alcoholism.

CDT TEST IN SERUM BY UV - FAST - MONOREAGENT

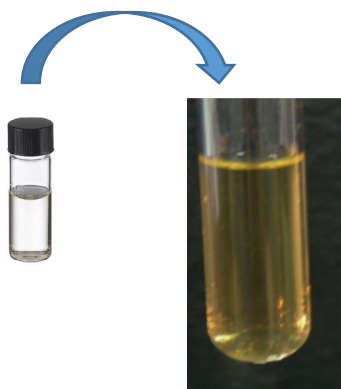
Z68210 (100 TESTS)

Z68215 (500 TESTS)



CDT TEST IN SERUM BY UV - FAST - MONOREAGENT

PRINCIPLE OF THE METHOD



Serum is complexed with an appropriate reagent



Centrifugation



Injection into HPLC



TECHNICAL REQUIREMENTS


Required

Binary HPLC System with loop of 50 μ l in peek.
Spectrophotometric Detector UV/VIS $\lambda=460$ nm
Chromatograms Recorder

Required

Autosampler
Operational Computer

Serum
collection

Dispense 3 ml of venous blood into a tube for serum without gel. Centrifuge at 4,000 rpm for 5 minutes. Separate the serum and store it at  -15°C .

It is stable at  -25°C for 4 weeks.

KIT CONTENT

NAME	DESCRIPTION
Reagent A	Complexing solution
Lyophilized Serum Control	
Reagent M1	Mobile Phase M1
Reagent M2	Mobile Phase M2
Reagent M3	Mobile Phase M3

ANALYTICAL PROCEDURE

100 μ l of **CONTROL**



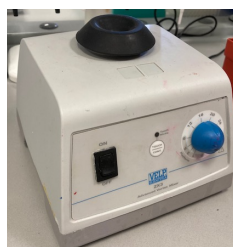
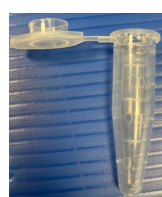
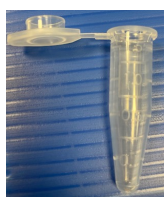
50 μ l reagent A
(Complexing Solution)



100 μ l of **SAMPLE**



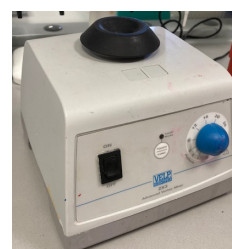
50 μ l reagent A
(Complexing Solution)



Vortex 10 seconds



Centrifuge, take the
supernatant and add
H₂O HPLC grade



Vortex 10 seconds

Injection into HPLC



TECHNICAL FEATURES

ABBREVIATIONS	MEANING
C LLOQ	Concentration at low limit of quantification
C1	Low concentration
C2	Median concentration
C3	High concentration
Max C	The highest concentration

ANALYTE	CONCENTRATIONS USED TO CALCULATE REPRODUCIBILITY AND ACCURACY (%)				
	C LLOQ	C1	C2	C3	MAX C
CDT	1.20	1.82	2.52	3.55	4.34

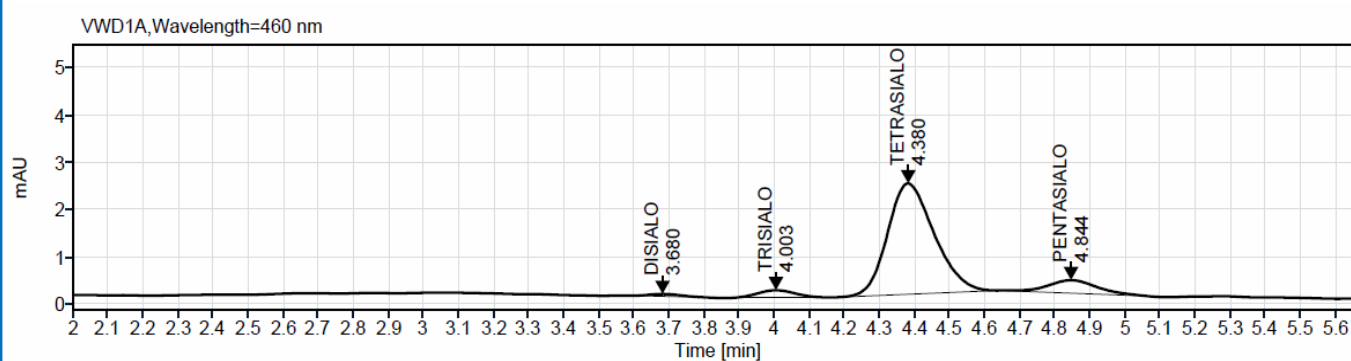
TECHNICAL FEATURES

ANALYTE	ACCURACY INTRA SERIE (RELATIVE ERROR %)		ACCURACY INTER SERIE (RELATIVE ERROR %)	
	C1	C3	C1	C3
CDT	3.66	3.10	5.92	3.72

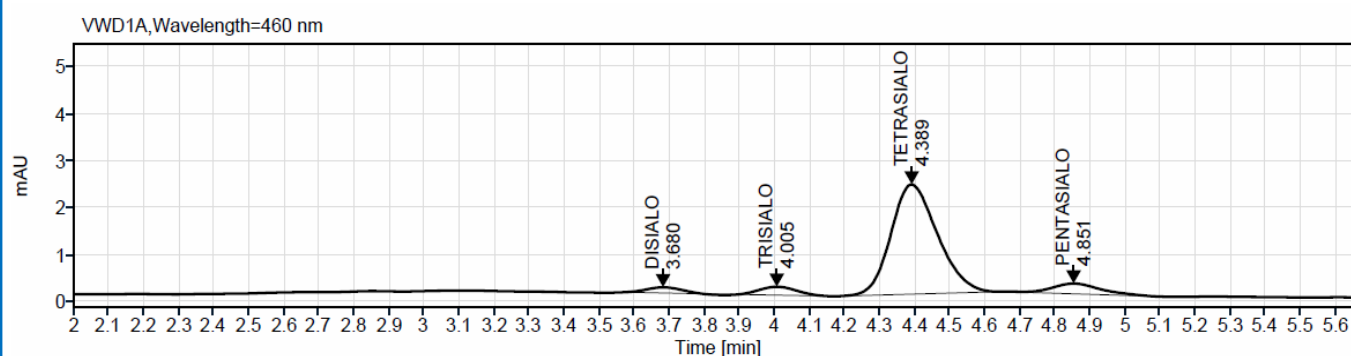
ANALYTE	REPRODUCIBILITY INTRA SERIE (CV%)			REPRODUCIBILITY INTER SERIE (CV%)		
	C LLOQ	C2	MAX C	C LLOQ	C2	MAX C
CDT	6.75	8.95	7.10	7.57	8.51	8.28

DETERMINATION OF THE DISIALO TRANSFERRIN

REFERENCE CHROMATOGRAMS



Control Level 1



Control Level 2

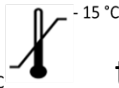
TRANSFERRIN	RETENTION TIME (min)
Disialo	3.68
Trisialo	4.00
Tetrasialo	4.39
Pentasialo	4.85

The reference chromatograms have been obtained by using HPLC Agilent 1260 infinity II and kindly provided by "CENTRO ANALISI NICOTRA S.A.S."

THE ADVANTAGES OF THE EUREKA KIT

Eureka kit, applied to the HPLC system for the determination of CDT in serum,

guarantees

- ✓ Straightforward and Quick Sample Preparation
- ✓ The serum sample volume required is only 100 µl
- ✓ No reconstituting reagent is needed
- ✓ A single reagent is dispensed
- ✓ Robust, Fast and Reliable CE-IVD method
- ✓ High reproducibility and great accuracy
- ✓ Time and Cost Optimization
- ✓ Stability for 3 years
- ✓ When opened and stored properly at  the shelf life of our internal standards and calibrators is 6 months
- ✓ Post sales service and training courses



The kit has been validated with the IFCC Standards that show good accuracy by integration with baseline.

Homogeneity of controls and calibrators has been determined according to and meets the criteria of ISO 13528.

BIBLIOGRAPHY

- [1] Dasgupta *et al.* "Alcohol and its Biomarkers", Science direct, 2015.
- [2] Stibler H "Abnormal micro-heterogeneity of transferrin in serum and cerebrospinal fluid in alcoholism", Acta Med Scand 1978;204:49-56
- [3] Stibler H, Borg S, Joustra M "A modified method for the assay of carbohydrate-deficient transferrin (CDT) in serum" Alcohol Alcohol Suppl 1991;451-4
- [4] Jeppsson J-O, Arndt T, Schellenberg F, Wielders JPM, Anton RF, Whitfield JB, Helander A "Toward standardization of carbohydrate-deficient transferrin (CDT) measurements: I. Analyte definition and proposal of a candidate reference method" Clin Chem Lab Med 2007;45:558-562



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