

## To Clot or Not To Clot: Are there Predictors of Clinically Significant Thrombus Formation in Patients with Gastroparesis and Prolonged IV Access?

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**Abstract** Some patients with gastroparesis (GP) require sustained central intravenous access for hydration, medication and/or nutrition, leaving them at risk for venous thrombosis. We studied a group of 53 patients with gastroparesis for identifiable risk factors of clinically significant thrombosis. Patients requiring prolonged central IV access fell into two groups: those who had clinical incidence of IV catheter-related thrombosis confirmed radiologically (CLOT,  $n = 14$ ), and those who did not form IV catheter thrombosis (NOCLOT,  $n = 39$ ). We analyzed and compared clinical symptoms, serum/plasma coagulation studies, and autoimmune antibodies in the CLOT and NOCLOT groups. Patients in the CLOT group had statistically more Scl 70 antibodies than did the NOCLOT group, and another autoantibody, Ku 66, was found in higher titers in the NOCLOT group than the CLOT group. Other autoimmune and coagulation factors were not statistically different between the two groups, although a subgroup of CLOT patients had lower plasma Protein S levels. We conclude that the presence of Scl 70 autoantibodies is associated with increased clotting risk in this group of GP patients, and that the Ku 66 antibody may be associated

with decreased risk of thrombosis in patients with GP. These findings, coupled with lower Protein S levels in some CLOT patients, suggests that autoimmune factors may be associated with GP patients who thrombose IV access versus patients who do not.

**Keywords** Gastroparesis · Thrombosis · Nausea · Vomiting

### Introduction

Gastroparesis also known as upper gastrointestinal luminal gut failure (LGF), is a syndrome that manifests as a variety of clinical symptoms, primarily nausea, vomiting, bloating, early satiety, anorexia, and abdominal pain. Patients with GP symptoms often have additional symptoms that overlap with other syndromes, such as migraine headaches, fibromyalgia, interstitial cystitis, endometriosis, and systemic hypercoagulability [1, 2]. A recent report of 60 consecutive patients revealed that over 90% of GP patients may harbor a measurable hypercoagulable state [1]. These patients frequently have central venous catheter (CVC) associated thrombosis and other clinically significant thrombotic events [3, 4].

Often, GP patients are refractory to available medications and require prolonged CVC access for hydration, nutrition, and parenteral medication. This poses the dilemma of sustained central venous access in the setting of an already hypercoagulable patient population [3]. Although many of the GP patient population have prothrombotic coagulopathies [1, 2, 5], not all patients form clinical thrombi despite the thrombophilic state. Since thrombotic events frequently complicate GP patients' clinical course and overall outcome [3, 6], predictors of

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clotting would be a helpful tool in the clinical management of gastroparetics.

We retrospectively evaluated a group of patients with GP and prolonged central venous access as two subsets: those who formed CVC associated thrombosis (designated CLOT group) and those who had no clinical incidence of thrombosis (designated NOCLOT group). We analyzed clinical data as well as serum/plasma coagulation and autoimmune markers for differences between the CLOT and NOCLOT groups. Our goal was to compare these two groups to determine if identifiable risk factors for clinical thrombosis development could be determined.

### Patients

We studied a group of 53 patients (9 males and 44 females, with a mean age of 42 years) with GP who all required CVC placement. One patient had post-viral LGF, seven had an underlying diagnosis of diabetes, and the remainder had idiopathic disease. Forty-nine were Caucasian, three were African American, and one was Hispanic. Fourteen of the 53 patients (26%) had clinical thrombosis related to their CVC and 39 had no clinical thrombosis. There was 1 male and 13 females in the CLOT group. The NOCLOT group was made up of 8 males and 31 females. These patients had all undergone extensive evaluations to determine possible underlying causes of the GP, including full-thickness small bowel biopsies in many patients. Of the 14 patients in the CLOT group, 10 had full-thickness small bowel biopsies, of which 1 patient had histological abnormalities of the gastric smooth muscle (myopathy), 5 had histological abnormalities of the enteric innervation (neuropathy), and 4 were normal. Of the 39 patients in the NOCLOT group, 26 patients had full-thickness small bowel biopsies, of which 3 had small bowel muscle abnormalities, 18 had enteric innervation abnormalities, and 5 were normal. Patients in the NOCLOT group reported a higher vomiting score than the CLOT group, yet the CLOT group reported higher incidence of nausea than the NOCLOT group (see Table 1).

CVC (IV) access, for the purposes of this review, was defined as any central IV access intended for use for medication and/or nutrition for at least 6 weeks. The CVC thromboses in these patients were defined as catheter associated DVT as opposed to catheter thrombotic occlusion. This distinction was made clinically and, in most cases, CVCs were in place for at least 1 week before CVC associated thrombosis was diagnosed. Each thrombosis was confirmed radiologically by either duplex ultrasound or venography.

**Table 1** Demographics, gastric biopsy, and clinical symptoms

	Clot ( <i>n</i> = 14)	No clot ( <i>n</i> = 39)
Male	1	8
Female	13	31
Average age	38.8	42.5
Full thickness biopsy	1 myopathy 5 neuropathy 4 normal <i>n</i> = 10	3 myopathy 18 neuropathy 5 normal <i>n</i> = 26
Nausea score	8.65 ± 0.98	8.27 ± 1.26
Vomiting score	6.53 ± 1.75	7.65 ± 1.28
TSS (total symptom score)	37.46 ± 10.01	36.58 ± 6.00
IDIOMS	17.81 ± 5.74	16.74 ± 4.07

### Methods

We examined differences in baseline GI symptoms and health related quality of life, coagulation studies, and autoimmune antibodies in the CLOT and NOCLOT groups. Patients were asked to rate their baseline symptoms on a 0–10 scale (0 = none, 1 = mildest, 10 = worst) in five areas: nausea, vomiting, bloating, abdominal pain, and anorexia. These were then totaled to calculate a Total Symptom Score (TSS); the maximum TSS possible was 50. The TSS was compared between the two groups. Health-related quality of life (HRQOL) was rated by an investigator-derived independent outcome measure (IDIOMS), a health resource measure as recently described [7] (see Table 1).

We also compared serum/plasma coagulation studies in the CLOT versus NOCLOT groups. We evaluated anticardiolipin antibodies, thrombin time, Protein S, and the Lupus anticoagulant. See Table 2 for assay methodologies.

The Western blot for autoantibodies (Immunovision, Springdale, Ark.) was used to test for various autoantibodies present in serum. The Western Blot (ANABLOT) was an enzyme-linked immunoassay intended for concurrent qualitative determination of human antibodies in serum with specificity to the following classically identified nuclear antigens: Scl 70, Scl 105, SSB 43, Sm 16, Sm 18, and Ku 66. The antigens had been electrophoresed and

**Table 2** Assay methodologies

Coagulation study	Assay
Anticardiolipin antibodies	ELISA
Thrombin time	Clot-endpoint testing
Protein S activity	Chromogenic substrate assay
Lupus anticoagulant	Dilute Russel Viper Venom time with hexagonal phase phospholipid confirmation

transferred to nitrocellulose with separation by molecular weight by the manufacture.

The ANABLOT procedure included equilibration of all reagents to room temperature for approximately 25 min. The ANABLOT strips were placed into individual troughs and labeled appropriately. A total of 2.5 ml of diluent buffer was added to each trough and 2.5 ml of serum added to the designated patient troughs and controls. The diluent and serum were mixed and incubated for 1 h at room temperature with gently continuous agitation on an orbiting table rotator. After incubation, each strip was washed by vacuum removal of fluid and 2.5 ml washing buffer was added, the troughs were then agitated for 1 min and incubated without agitation for 4 min. The wash was then repeated twice. Following the wash, 2.5 ml of working conjugate solution was added to each trough. The strips were then agitated gently for 1 h at room temperature. The strips were washed as before three times. The strips were then transferred to a clean trough and 5 ml of substrate/color development solution was added for each strip. The strips were incubated for 3–10 min to allow the colored bands to appear. The reactions were then stopped by rinsing each strip with distilled water. The strips were allowed to air dry and bands mapped according to manufacturer's guidelines. Positive and negative controls were run with each batch of strips. A positive band was determined when a 1+ or greater staining intensity was demonstrated.

In order to determine the overall immunologic reaction, a banding score was developed to standardize the bands present. The GI banding score (GIBS) was as follows: any band present that did not belong to a classic auto antibody pattern was awarded 1 point; any single band present of a classic autoantibody pattern was awarded 2 points; two bands present in any classic autoantibody pattern containing three or more bands was awarded 5 points; complete set of bands for any classic pattern containing two bands, or

complete set of bands for any classic pattern containing three bands, or three bands present for any classic pattern containing four bands were awarded 10 points; complete set of bands present for any classic pattern containing four bands was awarded 15 points. The total points were added to reflect the total autoantibody score as well as reporting of specific bands based on their classic patterns. The GIBS was based on the number and type of bands present and not the intensity of the individual bands. These were compared against standardized controls obtained from blood donors [8].

Results were compared by non-paired *t*-tests and reported as mean  $\pm$  standard error. Statistical significance was defined as a *P*-value less than or equal to 0.05. This review was approved by the University of Tennessee-Memphis IRB.

## Results

The baseline symptoms and IDIOMS were similar between the two groups, as reported above.

For the coagulation studies, our study found a marker that was different between the two groups. Lupus anticoagulant data showed that 100% of the CLOT group had normal levels. However, in the NOCLOT group, 10.2% had abnormally high levels of the Lupus anticoagulant. Protein S was lower in the CLOT group ( $64.35 \pm 4.64$  vs  $71.4 \pm 3.43$ ), but the results were not significantly different,  $P > 0.1$ . Also, a subgroup of 21 patients had Protein S deficiency, defined as  $<65\%$  of normal. In this subgroup, the averages for Protein S in the CLOT group ( $n = 6$  or 42%) was lower ( $47.0 \pm 2.96$  and  $50 \pm 2.66$ ) in the NOCLOT group ( $n = 15$  or 38%), but were not statistically significant. The averages of the CLOT versus the NOCLOT group for the other measures were similar: ACL

**Table 3** Coagulation studies

Test (data avail/total pts)	Clot ( $n = 14$ )	No clot ( $n = 39$ )	Reference range	<i>P</i> value
1 ACL antibodies (anticardiolipin) (50/53)	$8.07 \pm 1.98$ (12/14) 85%	$7.41 \pm 1.29$ (38/39) 97%	15–22.9	$>0.2$
2 Thrombin time (47/53)	$15.73 \pm 2.58$ (11/14) 79%	$14.39 \pm 0.57$ (36/39) 92%	16.2–21.8	$>0.2$
3 Protein S (53/53)	$64.35 \pm 4.64$ (14/14) 100%	$71.4 \pm 3.43$ (39/39) 100%	60–159	$>0.2$
4 Lupus anticoagulant (% with abnormal levels) (47/53)	0% (11/14) 78%	10.2% (36/39) 92%	0%	

**Table 4** Subgroup with Protein S deficiency

Test (data avail/total pts)	Clot ( $n = 6$ )	No clot ( $n = 15$ )	Reference range	<i>P</i> value
1 Protein S % (21/52)	$47 \pm 2.96$ (6/14) 42%	$50 \pm 2.66$ (15/39) 38%	60–159	$>0.2$
2 Lupus anticoagulant (21/52)	$0.66 \pm 0.21$ (6/14) 42%	$1.07 \pm 0.27$ (15/39) 38%	$<1.2$	0.13

**Table 5** ANABLOT and GIBS

	Test (data available/total points)	Clot ( <i>n</i> = 14)	No clot ( <i>n</i> = 39)	<i>P</i> value
1	GIBS (52/53)	11.23 ± 3.00 (13/14) 92%	14.56 ± 2.36 (39/39) 100%	0.34
2	Scl 70 (53/53)	0.74 ± 0.11 (14/14) 100%	0.28 ± 0.07 (39/39) 100%	0.04
3	Scl 105 (53/53)	0.21 ± 0.05 (14/14) 100%	0.43 ± 0.06 (39/39) 100%	0.14
4	SSB 43 (53/53)	0.35 ± 0.16 (14/14) 100%	0.58 ± 0.12 (39/39) 100%	0.27
5	Sm 16 (53/53)	0.07 ± 0.11 (14/14) 100%	0.07 ± 0.02 (39/39) 100%	0.94
6	Sm 18 (53/53)	0.14 ± 0.03 (14/14) 100%	0.02 ± 0.04 (39/39) 100%	0.26
7	Ku 66 (50/53)	0.07 ± 0.07 (14/14) 100%	0.47 ± 0.16 (36/39) 92%	0.04

antibodies  $8.07 \pm 1.98$  versus  $7.41 \pm 1.29$  ( $P > 0.2$ ), thrombin time  $15.73 \pm 2.58$  versus  $14.39 \pm 0.57$  ( $P > 0.2$ ) (see Tables 3 and 4).

For serum autoimmune studies, the average GIBS score for the CLOT group was lower ( $11.23 \pm 3.00$  and  $14.56 \pm 2.36$ ) than the NOCLOT group, but were not statistically significant. There were differences in two autoantibodies: The average Scl 70 for the CLOT group was higher ( $0.74 \pm 0.11$  and  $0.28 \pm 0.07$ ) versus the NOCLOT group ( $P < 0.05$ ) and the average Ku 66 for the CLOT group was lower ( $0.07 \pm 0.07$  and  $0.47 \pm 0.16$ ) than in the NOCLOT group. The averages of the individual autoantibody levels for the CLOT versus the NOCLOT groups were higher for Scl 105 ( $0.21 \pm 0.05$  vs  $0.43 \pm 0.06$ ,  $P = 0.14$ ) but were similar for SSB (43  $0.35 \pm 0.16$  vs  $0.58 \pm 0.12$ ;  $P > 0.2$ ); for Sm 16 ( $0.07 \pm 0.11$  vs  $0.07 \pm 0.02$ ;  $P > 0.2$ ) and Sm 18 ( $0.14 \pm 0.03$  vs  $0.02 \pm 0.04$ ;  $P > 0.2$ ) (see Table 5).

## Discussion

Our group of 53 patients represents a wide diversity of the patient population with gastroparesis. This study group included males and females and three different ethnicities and ages (range 9–87 years). All had drug-refractory gastroparesis and each had prolonged central venous access for medication and/or nutrition. The two patient groups (CLOT vs NOCLOT) stratified by clinical outcome were then compared by baseline symptom and laboratory parameters. There were no clinically significant differences in either group at baseline.

Our study found differences in the coagulation measures of lupus anticoagulant and a subgroup of protein S. The finding of a higher percentage of this patient group with abnormally elevated lupus anticoagulant measures is not surprising considering that many GP patients have other signs and symptoms of autoimmune diseases [9]. Also, the presence of low protein S is consistent with recent reports of pseudo-protein S which may exist in some of these patients.

Our study of autoimmune antibodies in these patients was intended to determine whether or not autoimmune factors are associated with thrombosis in this patient group. The Scl 70 autoantibody (and also the Scl 105 antibody, which was also elevated) is consistent with other reports of these antibodies in patients with gut neuro-muscular disorders [10]. The presence of lower Ku66 in the CLOT group is also worthy of comment, as this antibody is also found in several autoimmune diseases.

This study has applications to patient care as GP patients are at increased risk for thrombus formation, and any markers that might identify the patients at increased risk could have clinical usefulness. GP patients are often hospitalized for prolonged periods of time, frequently with IV access, which further increase their risk for clotting.

Drawbacks of our study were the small sample size ( $n = 52$ ) and the retrospective nature of the study. Future studies of thrombus formation in patients with the symptoms of gastroparesis should focus on prospective analysis.

## Conclusion

In this retrospective review of patients with the symptoms of gastroparesis and IV access, several factors were determined to be different from patients who clotted, or not.

A subgroup of protein S was found to be different between the CLOT and NOCLOT groups. We also conclude that higher levels of the Scl 70 antibody are found in GP patients who have clinical thrombosis. Conversely, the Ku 66 antibody is present in higher levels in GP patients who do not form complicating thromboses. Since serum autoimmune analysis is easily and inexpensively performed, it may have use as a screening tool for motility patients who require IV access. Future prospective studies to develop a protocol for screening GP patients with IV access for increased risk of thrombosis may want to include both autoimmune and coagulation studies.

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## References

1. Blanchard K, Rock W, Schmiege R, Araghizadeh F, Borman K, Minocha A, Abell TL (2004) Hypercoagulability is overwhelmingly present (93%) in patients with severe gastroparesis. *Gastroenterology* 126:A-374
2. Lohbrano A, Blanchard K, Abell TL, Minocha A, Rock W (2005) Hypercoagulability is found in a remarkably high percentage (89%) of patients with severe gastroparesis. *J Investig Med* 53:S273
3. Kumar A, Lomas R, Abell T, Dugdale M, Voeller G, Smalley D (1994) Serum autoantibodies predispose to superior venacaval syndrome associated with central access in gastroparesis. *Gastroenterology* 106:A528
4. Alkheshen M, Edick C, Hak L, Ismail MK, Nash K, Deither S, Graves PA, Dugdale M, Smalley D, Abell T (2000) High incidence of protein S abnormalities in patients with catheter-related thrombosis. *Gastroenterology* 118:1064
5. Duncan U, Abell T, Smalley D, Kuns J, Van Frank T, Dean P (1994) Autoantibodies in chronic dyspepsia: prevalence and possible diagnostic role. *Gastroenterology* 106:A491
6. Werkman R, Smalley D, Duncan U, Dugdale M, Taylor J, Kumar A, Abell TL, Voeller G (1995) Is thrombosis of central venous access in idiopathic upper GI dysmotility related to the presence of circulating autoantibodies? *Gastroenterology* 108:A734
7. Cutts TF, Luo J, Starkebaum W, Rashed H, Abell TL (2005) Is gastric electrical stimulation superior to standard pharmacologic therapy in improving GI symptoms, healthcare resources, and long-term healthcare benefits? *Neurogastroenterol Motil* 17:34–35
8. Shanklin DR, Smalley DL (2002) Pathogenetic and diagnostic aspects of siliconosis. *Rev Environ Health* 17:85–105
9. DeGiorgio R, Guerrini S, Barbara G, Stanghellini V, DePonti F, Corinaldesi R, Moses PL, Sharkey KA, Mawe GM (2004) Inflammatory neuropathies of the enteric nervous system. *Gastroenterology* 126:1872–1883
10. Duncan U, Abell TL, Kuns J, Waters B, Voeller G, Dean P, Smalley D (1994) Serum autoimmune antibody levels as predictors of morphologic abnormalities of nerve and gut. *Gastroenterology* 106:A491
11. Kumar A, Abell TL, Voeller G, Werkman R, Smalley D, Duncan U, Dugdale M, Taylor J (1995) Is thrombosis of central venous access in idiopathic upper GI dysmotility related to the presence of circulating auto-antibodies. *Gastroenterology* 108:A734. Presented, AGA National Meeting, San Diego, May 1995
12. Bhaskar SK, Abell TL, Smalley D, Familoni L, Voeller G (1996) Correlation of full thickness biopsies and serum autoantibody scores in patients with unexplained nausea and vomiting. *Gastroenterology* 110:A634
13. Idris A, Nash K, Smalley D, Voeller G, Seitcher S, Dugdale M, Abell TL (1998) Correlation between the occurrence of thrombosis and serum autoantibody levels in luminal gut failure patients with central IV access. (DDW, May 15–22. New Orleans, LA)
14. Alkheshen MA, Edick C, Hak L, Ismail MK, Nash K, Deitcher S, Graves PA, Dugdale M, Smalley D, Abell TL (2000) Patients with IV catheter related thrombosis have multiple coagulation and autoimmune abnormalities on baseline diagnostic tests. *Gastroenterology* 118:A1331