AppNote 1/2010

Full Automation of ASTM Method D6584-07 “Standard Test Method for the Determination of Free and Total Glycerin in B-100 Biodiesel Methyl Esters by Gas Chromatography” using a GERSTEL Dual Rail PrepStation

John R. Stuff, Jacqueline A. Whitecavage
Gerstel, Inc., 701 Digital Dr. Suite J,
Linthicum, MD 21090, USA

KEYWORDS
Biodiesel, Automation, ASTM D6584-07

ABSTRACT
Biodiesel is defined as mono-alkyl esters of long chain fatty acids derived from vegetable oils or animal fats which conform to ASTM D6751 specifications for use in diesel engines. Biodiesel is produced from the fat or oil by transesterification. Glycerin is an unwanted byproduct of the reaction and must be removed from the final product.

ASTM Method D6584-07 measures the amount of residual free and bonded (mono-, di-, and triglycerides) glycerin in the biodiesel fuel. Standard and sample preparation, which requires a derivatization step, can be laborious and time consuming.

Using the GERSTEL MPS 2 Dual Rail PrepStation and MAESTRO control software, the entire method, including standards and sample preparation, derivatization, dilution, and analysis can be fully automated. This paper shows the details of the automation.

Introduction

A dual rail PrepStation was set up to fully automate the procedure outlined in ASTM D6584-07 for the determination of free and total glycerides in B-100 biodiesel fuel. The goal is to provide complete automation of standard preparation, automate next sample preparation within the time frame of the GC run, and provide other user-friendly features of the Maestro software control for the ASTM method. Full automation frees up the analyst's time for other laboratory operations.

Experimental

Instrumentation. Analyses were performed on an Agilent Technologies 6890 GC with flame ionization detector (FID), PTV inlet (CIS 4, GERSTEL) and Dual Rail MPS 2 robotic sampler with 10 µL on-column syringe and an 80 µL sideport syringe with diluter module (GERSTEL).

Analysis conditions.

PTV:	On-Column 60°C (0.05 min); 0.2°C/sec; 230°C (2 min); 0.5°C/sec; 380°C (10 min)
Column:	10 m Rtx-Biodiesel TG (Restek) d_i = 0.32 mm d_f = 0.1 µm with 2m integrated guard column, d_i = 0.53 mm
Pneumatics:	He, constant flow = 3 mL/min
Oven:	50°C (1 min); 15°C/min; 180°C; 7°C/min; 230°C; 30°C/min; 380°C (10 min)
FID:	380°C

Standard description. A biodiesel standards kit (#D6584-SS), containing separate 5 mL ampoules of 500 ng/µL glycerin, 5000 ng/µL monoolein, 500 ng/µL diolein, 5000 ng/µL triolein, 1000 ng/µL butanetriol, and 8000 ng/µL tricaprin, all in pyridine, was purchased from Analytical Services, Inc. (The Woodlands, TX). The kit also contained MSTFA and heptane. Biodiesel B-100 was purchased locally.

Sample preparation. The glycerin, mono-, di- and triolein standards were placed in 2 mL screw cap vials in Positions 1-4, respectively, of a VT-98 tray. The butanetriol, tricaprin and MSTFA were put in 10 mL vials with PTFE snap caps and placed in Positions 3, 4, and 5 in the 5 position 10 mL wash tray. Empty 10 mL screw cap vials were placed in Positions 1-5 of the VT-32-10 tray. Biodiesel B-100 samples, approximately 100 mg, were weighed directly into 10 mL screw cap vials and placed on the VT-32-10 tray. The samples and standards were prepared using a GERSTEL MAESTRO PrepSequence.

Sample introduction. The samples were prepared in vials placed in the VT32-10 tray. One microliter of the standard or sample was injected into the CIS 4 using the MPS 2 autosampler with a 10 µL liquid syringe with replaceable 26 gauge needle for on-column injection

Results and Discussion

The GC system used for this study is shown in Figure 1. The method calls for the preparation of a five point calibration curve for glycerin, mono-, di-, and triolein from the stock standards. The required volumes are shown in Table 1.

Figure 1. System setup.

Table 1. Required stock standard volumes.

Standard Number	1	2	3	4	5
µL Glycerin Stock	10	30	50	70	100
µL Monoolein Stock	20	50	100	150	200
µL Diolein Stock	10	20	40	70	100
µL Triolein Stock	10	20	40	70	100

The GERSTEL MAESTRO PrepBuilder function is used to automate the preparation and injection of standards and samples. An example of a standard preparation loop for the addition of the diolein stock to the five standard vials is shown below. The individual steps are selected by mouse-click from a drop-down menue.

ACTION	METHOD / VALUE	SOURCE	DESTINATION
PREP Vials 3-3	Extensive		
(R) ADD	Add 10 uL Std	3 @ Tray2,VT98	1 @ Tray1,VT32-10
(R) ADD	Add 20 uL Std	3 @ Tray2,VT98	2 @ Tray1,VT32-10
(R) ADD	Add 40 uL Std	3 @ Tray2,VT98	3 @ Tray1,VT32-10
(R) ADD	Add 70 uL Std	3 @ Tray2,VT98	4 @ Tray1,VT32-10
(R) ADD	Add 50 uL Std	3 @ Tray2,VT98	5 @ Tray1,VT32-10
(R) ADD	Add 50 uL Std	3 @ Tray2,VT98	5 @ Tray1,VT32-10
(R) DILUTE	Dilute Wash Syringe		Waste
END			

Samples are prepared by weighing approximately 100 mg into an 10 mL empty vial. The preparation of standards and samples is performed as follows:

1. Add 100 µL each of 2 Internal Standards
2. Add 100 µL Derivatizing Reagent, Mix, Wait 15 min
3. Dilute with 8 mL heptane, Mix
4. Analyze using a 1 µL on-column Injection

An example for the preparation and analysis loop of the five standards and five samples is shown below.

ACTION	METHOD / VALUE	SOURCE	DESTINATION
PREP Vials 1-10	Extensive		
(R) ADD	ADD First 50 uL	SolvRes1	Tray1,VT32-10
(R) ADD	ADD Second 50 uL	SolvRes1	Tray1,VT32-10
(R) DILUTE	Dilute Wash Syringe		Waste
(R) ADD	ADD First 50 uL	SolvRes2	Tray1,VT32-10
(R) ADD	ADD Second 50 uL	SolvRes2	Tray1,VT32-10
(R) DILUTE	Dilute Wash Syringe		Waste
(R) ADD	ADD First 50 uL	SolvRes3	Tray1,VT32-10
(R) ADD	ADD Second 50 uL	SolvRes3	Tray1,VT32-10
(R) DILUTE	Dilute Wash Syringe		Waste
(R) MOVE		Tray1,VT32-10	Agitator,AgiTray
(R) MIX	Mix 1 Minute		
(R) MOVE		Agitator,AgiTray	Tray1,VT32-10
(R) WAIT	15.00 min		
(R) DILUTE	Dilute 8 mL Heptane		Tray1,VT32-10
(R) MOVE		Tray1,VT32-10	Agitator,AgiTray
(R) MIX	Mix 1 Minute		
(R) MOVE		Agitator,AgiTray	Tray1,VT32-10
(L) INJECT	ASTM_D6584.m\maestro.mth	Tray1,VT32-10	Rear
END			

The first 2 ADD steps correspond to the addition of 100 µL of the butanetriol internal standard. After the syringe is washed, 100 µL of tricaprin internal standard is added, followed by another wash and 100 µL of the MSTFA to derivatize the sample/standard. The sample or standard is then mixed, a 15 minute wait step is inserted to allow the derivatization to be completed, heptane is added for dilution, followed by mixing and GC analysis.

The inject action step contains all the Agilent Chemstation and GERSTEL MPS 2 functions required for analysis of the samples. When integrated into Agilent ChemStation or MS ChemStation, one method and one sequence table runs the complete system.

Figure 2 shows a typical chromatogram obtained for Standard #3. The standard and internal standard peaks are labeled in the chromatogram. Table 2 shows the linear regression results for the standard curves. Good correlation is achieved for all standards. Figure 3 shows the calibration curve for diolein.

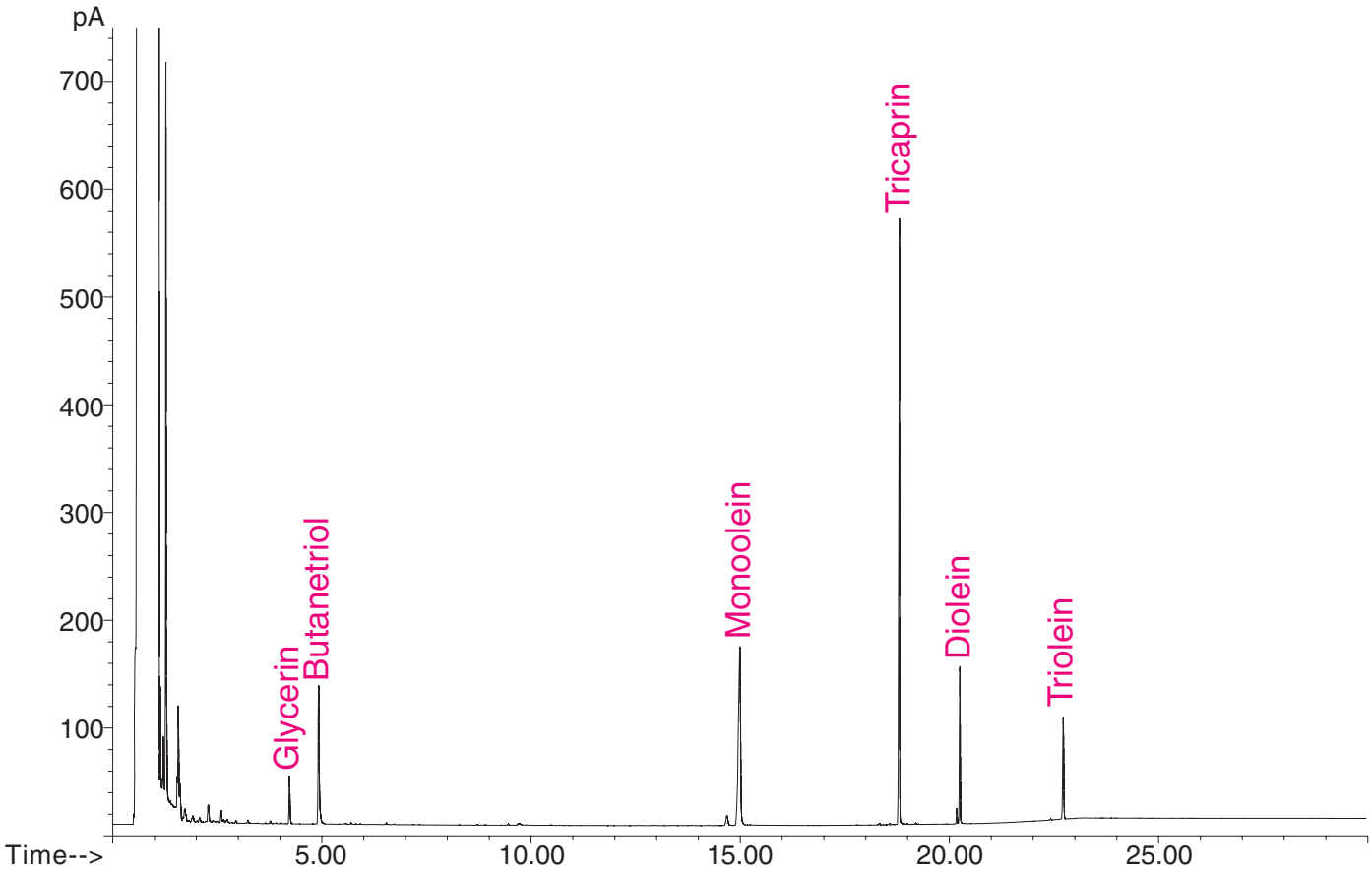

Figure 2. Chromatogram for standard #3.

Table 2. Regression data for standards.

Compound	r^2	m	b
Glycerin	0.9999	1.181	-0.0008
Monoolein	0.9997	1.247	-0.0122
Diolein	0.9987	1.0203	-0.0067
Triolein	0.9985	0.8946	-0.0120

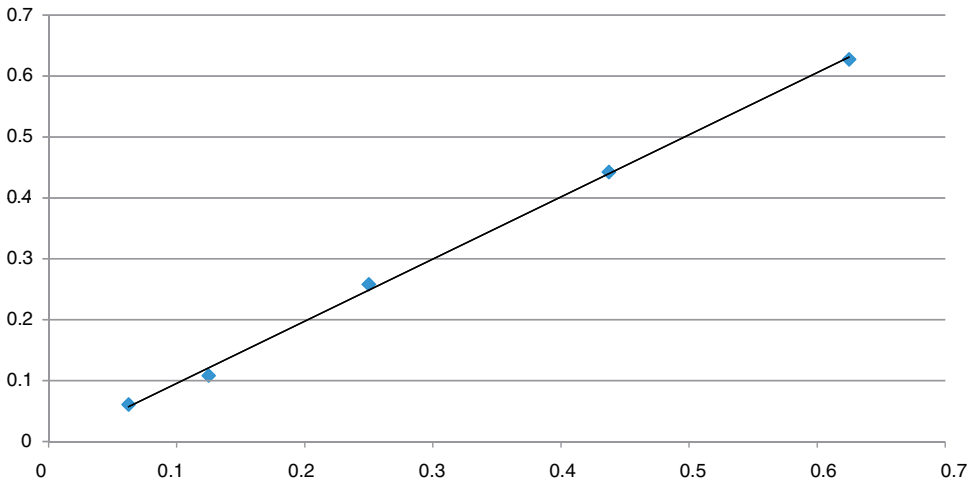

Figure 3. Diolein calibration curve.

Table 3 shows the results for the sample replicates (n=5). Reproducibilities are good with %RSDs ranging from 2.1-3.4%. A typical chromatogram for this sample type is shown in Figure 4.

Table 3. Mass percentages for B-100 Biodiesel.

	Glycerin	Monoglycerides	Diglycerides	Triglycerides
Sample 1	0.02608	1.107	0.1898	0.08909
Sample 2	0.02530	1.055	0.1864	0.09379
Sample 3	0.02662	1.059	0.1890	0.09398
Sample 4	0.02706	1.056	0.1858	0.09565
Sample 5	0.02670	1.071	0.1791	0.08861
Average	0.02635	1.070	0.1860	0.09220
Std Dev	0.000683	0.0220	0.00421	0.00316
%RSD	2.59	2.06	2.26	3.42

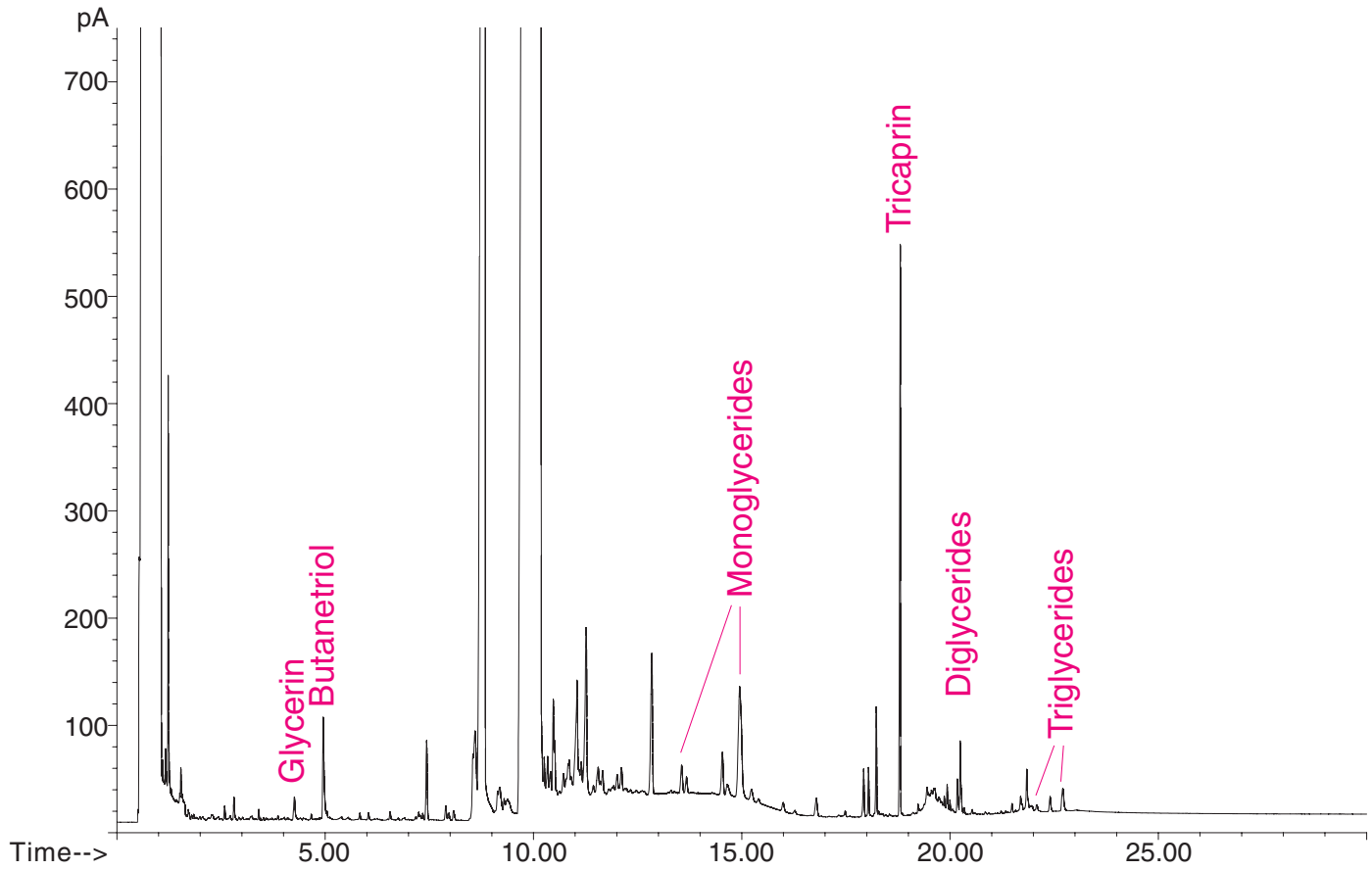

Figure 4. Chromatogram for biodiesel sample.

The preparation of each sample was completed in approximately 27 minutes using MAESTRO PrepBuilder functions, well within the total GC cycle time of 38 minutes (31 minute GC run + 7 minute cooldown). This means that samples can be prepared during the GC run of the preceding sample without adding to the overall analysis time, a fact which is clearly illustrated by the MAESTRO Sequence Scheduler window (Figure 5). The scheduler graphically illustrates that the sample prep and GC run segments of the analysis are performed in parallel, while giving a clear overview of the total analysis time needed in order to facilitate laboratory workflow planning.

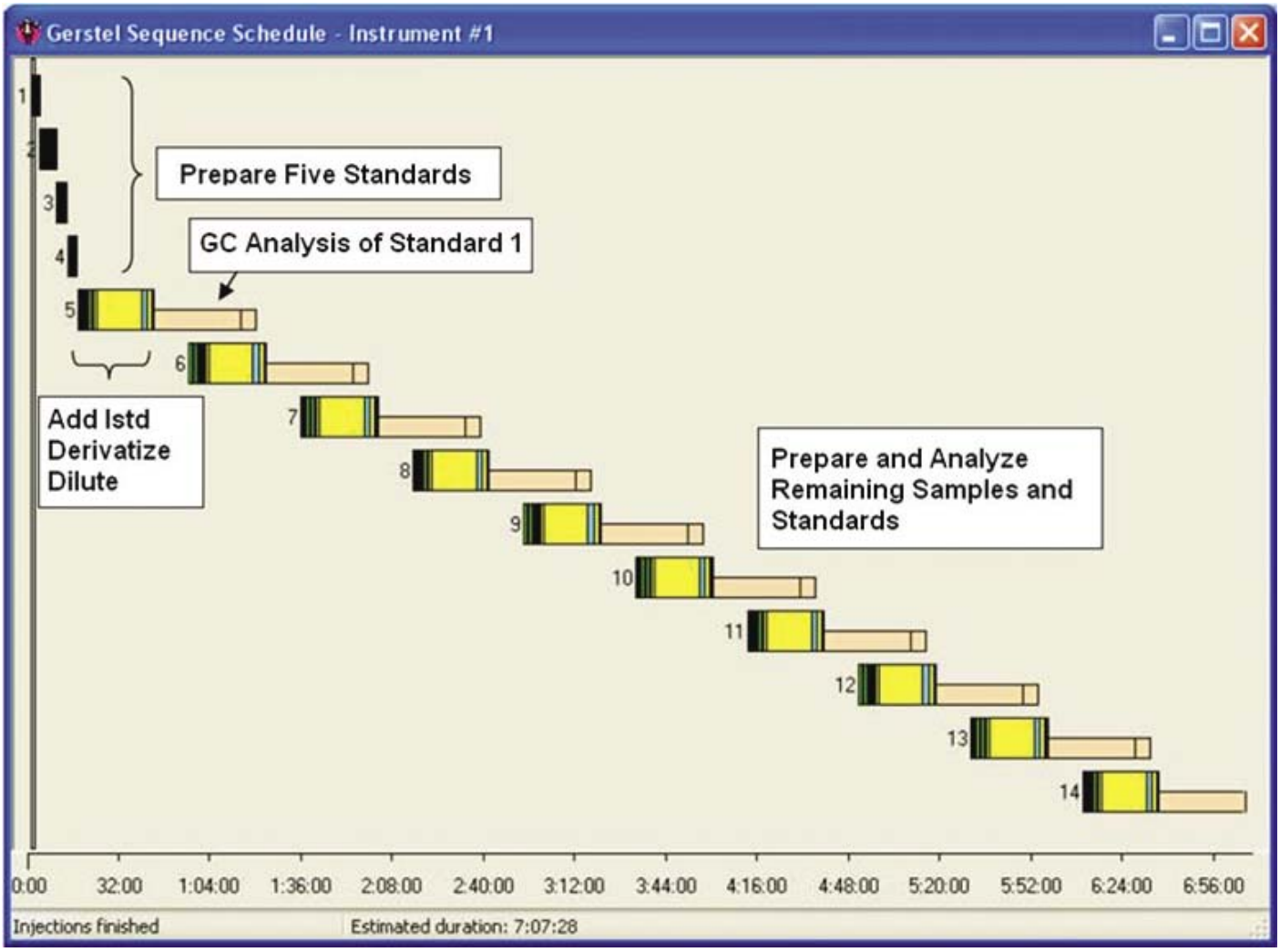

Figure 5. MAESTRO sequence scheduler

CONCLUSIONS

A dual rail PrepStation was programmed to prepare standards and samples, and inject samples using GERSTEL MAESTRO software. The results showed good linearity for the standards and good reproducibility (%RSDs from 2.1-3.4%, n=5) for the B-100 biodiesel samples. Complete automation of ASTM D6584-07 was accomplished with next sample preparation within the timeframe of the GC run.

GERSTEL GmbH & Co. KG
Eberhard-Gerstel-Platz 1
45473 Mülheim an der Ruhr
Germany
+49 (0) 208 - 7 65 03-0
+49 (0) 208 - 7 65 03 33
gerstel@gerstel.com
www.gerstel.com

GERSTEL Worldwide

GERSTEL, Inc.
701 Digital Drive, Suite J
Linthicum, MD 21090
USA
+1 (410) 247 5885
+1 (410) 247 5887
sales@gerstelus.com
www.gerstelus.com

GERSTEL AG
Wassergrabe 27
CH-6210 Sursee
Switzerland
+41 (41) 9 21 97 23
gerstelag@ch.gerstel.com
www.gerstel.ch

GERSTEL K.K.
1-3-1 Nakane, Meguro-ku
Tokyo 152-0031
SMBC Toritsudai Ekimae Bldg 4F
Japan
+81 3 5731 5321
+81 3 5731 5322
info@gerstel.co.jp
www.gerstel.co.jp

GERSTEL LLP
Level 25, North Tower
One Raffles Quay
Singapore 048583
+65 6622 5486
+65 6622 5999
SEA@gerstel.com
www.gerstel.com

GERSTEL Brasil
Av. Pascoal da Rocha Falcão, 367
04785-000 São Paulo - SP Brasil
+55 (11)5665-8931
+55 (11)5666-9084
gerstel-brasil@gerstel.com
www.gerstel.com.br

Awarded for the
active pursuit of
environmental sustainability