The number of biological/biomedical applications that require AMS to achieve their goals is increasing, and so is the need for a better understanding of the physical, morphological, and structural traits of high quality of AMS targets. The metrics of quality included color, hardness/texture, and appearance (photo and SEM), along with FT-IR, Raman, and powder X-ray diffraction spectra that correlate positively with reliable and intense ion currents and accuracy, precision, and sensitivity of fraction modern ($F_m$). Our previous method produced AMS targets of gray-colored iron–carbon materials (ICM) 20% of the time and of graphite-coated iron (GCI) 80% of the time. The ICM was hard, its FT-IR spectra lacked the sp$^2$ bond, its Raman spectra had no detectable G′ band at 2700 cm$^{-1}$, and it had more iron carbide ($\text{Fe}_3\text{C}$) crystal than nanocrystalline graphite or graphitizable carbon (g-C). ICM produced low and variable ion current whereas the opposite was true for the graphitic GCI. Our optimized method produced AMS targets of graphite-coated iron powder (GCIP) 100% of the time. The GCIP shared some of the same properties as GCI in that both were black in color, both produced robust ion current consistently, their FT-IR spectra had the sp$^2$ bond, their Raman spectra had matching D, G, G′, D+G, and D″ bands, and their XRD spectra showed matching crystal size. GCIP was a powder that was easy to tamp into AMS target holders that also facilitated high throughput. We concluded that AMS targets of GCIP were a mix of graphitizable carbon and Fe$\text{C}$ crystal, because none of their spectra, FT-IR, Raman, or XRD, matched exactly those of the graphite standard. Nevertheless, AMS targets of GCIP consistently produced the strong, reliable, and reproducible ion currents for high-throughput AMS analysis (270 targets per skilled analyst/day) along with accurate and precise $F_m$ values.

Graphitization of solid carbon of biological/biomedical origin is a two-step process: first the carbon of a sample of interest is oxidized to $\text{CO}_2$, and second, the $\text{CO}_2$ is then reduced to graphitic materials. Graphite is a well-ordered crystal structure of carbon that is classified as natural or synthetic.$^{1,2}$ The H$_2$ or Zn reduction method has long been used for carbon dating and more recently for biological/biomedical AMS applications.$^{3-9}$ A variety of terms such as amorphous carbon (a-C),$^{10}$ graphite,$^{5}$ a fullerene “graphite”,$^6$ or solid fullerene$^{11}$ have been used within and between prior reports to describe the material produced during the reduction of $\text{CO}_2$, CO, or both. While the physical, morphological, and structural characteristics of the AMS targets that had been produced seemed to vary within and between reports, they have not been studied systematically nor has the importance of their physical, morphological, and structural characteristics in maximizing ion current been well determined. Using Zn as the reductant, Jull et al.$^{12}$ first reported the formation of a mix of well-crystallized materials, poorly crystallized materials, and on occasion, a metal carbide (MeC). While nickel formed a different (more complicated) MeC compared to cobalt or iron,$^4$ MeC, in general, failed to produce a robust ion current so the ratios of $^{14}\text{C}/^{13}\text{C}$ varied by as much as 10% among replicate analyses.$^5$

Therefore, in the present paper, we describe the color, hardness, texture, scanning electron microscopy (SEM), FT-IR transmission, Raman spectrometry, and XRD analyses of the accelerator mass spectrometry (AMS) targets (carbon material that coated over the −400 mesh spherical iron powder, −400MSIP) as prepared by our previous$^7$ and optimized$^{13}$ methods. Our goal

(1) Oya, A.; Marsh, H. J. Mater. Sci. 1982, 17, 309–322.
(2) Wisaler, M. J. Power Sources 2006, 156, 142–150.
(3) Vogel, J. S.; Southon, J. R.; Nelson, D. E.; Brown, T. A. Nucl. Instrum. Methods Phys. Res., Sect. B 1984, 5, 289–293.
(4) Vogel, J. S.; Southon, J. R.; Nelson, D. E. Nucl. Instrum. Methods Phys. Res., Sect. B 1987, 29, 50–56.
(5) Vogel, J. S. Radiocarbon 1992, 34, 344–350.
(6) Ognibene, T. J.; Bench, G.; Vogel, J. S.; Peaseke, G. F.; Murov, S. Anal. Chem. 2003, 75, 2192–2196.
(7) Getachew, G.; Kim, S. H.; Burri, B. J.; Kelly, P. B.; Haack, K. W.; Ognibene, T. J.; Buchholz, B. A.; Vogel, J. S.; Modrow, J.; Clifford, A. J. Radiocarbon 2006, 48, 325–336.
(8) Lin, Y.; Dueker, S. R.; Follett, J. R.; Fadel, J. G.; Arjomand, A.; Schneider, F. D.; Miller, J. D.; Green, R.; Buchholz, B. A.; Vogel, J. S.; Phair, R. D.; Clifford, A. J. Am. J. Clin. Nutr. 2004, 80, 680–691.
(9) Clifford, A. J.; de Moura, F. F.; Ho, C. C.; Chuang, J. C.; Follett, J.; Fadel, J. G.; Novotny, J. A. J. Am. J. Clin. Nutr. 2006, 84, 1430–1441.
(10) Andree, M.; Beer, J.; Oeschger, H. B. G.; Hofmann, H. J.; Morenzoni, E.; Nessi, M.; Suter, M.; Wülfli, W. Nucl. Instrum. Methods Phys. Res., Sect. B 1984, 5, 274–279.
(11) Vogel, J. S.; Love, A. H. Methods Enzymol. 2005, 402, 402–422.
(12) Jull, A. J. T.; Donahue, D. J.; Hatheway, A. L.; Linick, T. W.; Toolin, L. J. Radiocarbon 1986, 28, 191–197.
(13) Kim, S. H.; Kelly, P. B.; Clifford, A. J. Anal. Chem. 2008, 80, 7651–7660.
was to identify the traits of AMS targets that produced intense and reliable ion currents for accurate, precise, reliable, and high throughput for biological/biomedical applications of AMS.

**EXPERIMENTAL SECTION**

**Reagents.** The reagents were the same as previously described except for the following: Potassium bromide (KBr, CAS No. 7758–02–3, ≥99%, FT-IR grade) and graphite standard (GST, CAS No. 7782–42–5, <20 μm, synthetic powder) were from Sigma-Aldrich (St. Louis, MO).

**AMS Target Preparation.** AMS targets were prepared by our previous method as well as by our optimized method for which we built a new heating block with 7 rows, 14 holes apiece, to which the KBr and GST were added. The interplane distance of crystallite to direction (measured in radians). The average crystal size was calculated using the Scherrer equation, \( t = \frac{Kλ}{B \cos Θ} \), where \( t \) is the averaged dimension of crystallines (or \( Lc \), stacking height of crystallite to \( c \)-direction), \( K \) is the Scherrer constant whose value is \( ~0.9 \) (a somewhat arbitrary value in the range 0.87–1.0), \( λ \) is the wavelength of X-ray radiation, and \( B \) is the half-width height of the diffraction intensity distribution (measured in radians).

**RESULTS**

Figure 1 is a carbon classification by which AMS targets could be characterized in a relative way. The classification enabled us to rank the degree of crystal (high to low) as graphite crystal > t-C > g-C > i-C, the i-C is highly disordered and approximates amorphous carbon (a-C). The isotropic carbon (i-C), graphitize semifdermer carbon (g-C), turbostratic carbon (t-C), and graphite panels were from a previous publication, with permission. Figure 2 summarized an overview of the second step of AMS target preparation using our previous and optimized methods. The left panel shows the setup and appearance of the −400MSIP and Zn dust before the reduction. The center and right panels show what happened to the −400MSIP and Zn dust during the reduction using our previous and optimized methods, respectively. Prior to the CO₂ reduction step, the Zn dust and the −400MSIP consisted of loose spherical particles (left panel). After reduction by our previous method (center panel), the Zn dust appeared as a fibrous (rubbery) Zn band-1 at 260 °C, a plasticized or plastic-like Zn band-2 at 380 °C, and two Zn mirrors (mirror 1 at 480 °C and mirror 2 at 525 °C), which appeared to be metallic Zn on the inner wall of the septa-sealed vial. Furthermore, the remaining Zn dust appeared as a Zn cake stuck to the bottom of the septa-sealed vial. Finally, the AMS targets appeared at 540 °C as a graphite-coated iron (GCI) fuzz or a featureless ICM on the −400MSIP at the bottom of the borosilicate inner vial. After reduction by our optimized method, the Zn dust was deformed and appeared as a thinner (compressed) and more fibrous Zn band at 240 °C, a Zn mirror at 380 °C, and a softer Zn cake with less sinter at 500 °C that stuck to the bottom of the septa-sealed vial. The AMS targets appeared as a graphite-coated Fe powder (GCIP) fuzz on the −400MSIP at the bottom of the borosilicate inner vial at 500 °C.

Table 1 summarized the physical and morphological characteristics of the GST as purchased from Sigma-Aldrich and −400MSIP as purchased from Sigma-Aldrich. It also summarized the physical and morphological characteristics of AMS targets of iron-carbon materials (ICM) (produced 20% of the time) and GCI (produced 80% of the time) by our previous method and of GCIP (produced 100% of the time) by our optimized method. The materials were ranked by the lightness of their color as −400MSIP > ICM > GST = GCI = GCIP. The ICM was four times harder than the GCI (\( p < 0.0001 \)); hardness of powders was not measurable. The photos showed that only the GST, −400MSIP, and GCIP were powders. In comparing SEM of GST to that of −400MSIP, GST was graphite sheets, while −400MSIP was

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(14) Gil, M. I.; Holcroft, D. M.; Kader, A. A. J. Agric. Food Chem. 1997, 45, 1662–1667.

(15) Mitcham, B.; Cantwell, M.; Kader, A. A. Perishables Handling News. 1996, 85, 1–5.

(16) Sadezky, A.; Muckenhuber, H.; Grothe, H.; Niessner, R.; Pöschl, U. Carbon 2005, 43, 1731–1742.

(17) Stanjek, H.; Häusler, W. Hyperfine Interact. 2004, 154, 107–119.

(18) Cullity, B. D. Elements of X-Ray Diffraction, 2nd ed.; Addison Wesley: Reading, MA, 1978.
spherical, and the particle size of each was \( \pm 37 \) \( \mu m \). The SEMs also showed the surface of ICM as featureless, of GCIP as uniform fuzz, and of GCI as nonuniform fuzz. Even though the SEMs alone did not distinguish between Fe<sub>3</sub>C crystal, i-C, g-C, t-C, and graphite.

**Figure 1.** Schematic of carbon classification based on graphite. The classification enabled the degree of crystal to be ranked (high to low) as graphite crystal > t-C > g-C > i-C. The i-C is highly disordered and approximates amorphous carbon (a-C). The isotropic carbon (i-C), graphitizable semiorder carbon (g-C), turbostratic carbon (t-C), and graphite panels were from a previous publication.\(^1\) Rhombohedral graphite panel was from http://www.bas.bg/cleps/events/see/Presentations/SED_07_Trifonova.pdf.

**Figure 2.** Summary of the temperature gradient along the septa-sealed vial using our previous\(^7\) (center panel) and our optimized\(^13\) (right panel) methods. It also shows SEMs of the Zn deposits along the septa-sealed vials and of the AMS targets (GCI, GCIP) in the borosilicate inner vials. Our previous method produced AMS targets of gray-colored ICM 20% of the time and of GCI 80% of the time; neither target was a powder. Our optimized method\(^13\) produced AMS targets of 100% GCIP 100% of the time.\(^13\) AMS targets of GCIP produced more intense and reliable ion currents and more accurate \( F_m \) values than those of ICM or GCI. The left and center panels were from ref 13. The SEM picture (third from top, GCIP) in the right panel was also from ref 13.
crystal, the carbon classification in Figure 1, enabled the degree of crystal to be ranked (high to low) as graphite crystal > t-C > g-C > i-C. The i-C is highly disordered and approximates a-C. Finally, the GCIP was the most facile to tamp into the AMS target holder.

Figure 3 showed FT-IR transmission spectra of GST and of the AMS targets ICM, GCI, and GCIP, all showed a water band at $\sim 1630$ cm$^{-1}$, and except for ICM, and all showed a band at $\sim 1580$ cm$^{-1}$ that corresponded to the sp$^2$ bond (C=C bond stretching) in graphite. The band at $\sim 1580$ cm$^{-1}$ indicated that GCI and GCIP each had a carbon hexagonal structure.

Figure 4 summarized the Raman spectra of GST, ICM, GCI, and GCIP (left panel) in relation to Raman spectra from the scientific literature (right panel) with permission. The D, G, and G’ (G’ = 2D = D$'$) bands correspond to the sp$^2$ bond (diamond-like carbon), sp$^3$ bond (graphite and graphite-like carbon), and stacking arrangement without disorder (graphite crystal), respectively. The GST showed intense G and G’ bands at 1566.3 and 2705.4 cm$^{-1}$ and weak D and D’ bands at 1339.0 and 3228.5 cm$^{-1}$, respectively. The ICM appeared to have only weak D and G bands at 1329.4 and 1584.7 cm$^{-1}$ and no evidence of G’ or a G + D combination band that would indicate second-order Raman scattering. The GCI showed intense D and G bands at 1321.5 and 1585.7 cm$^{-1}$, respectively. In addition, GCI showed G’, G + D combination, and D’’ bands at 2655.7, 2917.2, and 3203.6 cm$^{-1}$ respectively. The Raman spectrum of GCIP was similar to that of GCI, and it also showed the first-order (D and G bands) and second-order Raman scattering (G’, G + D, and D’’ bands). The full on-width half-maximum height (fwhm, $\Delta$) of D, G, and G’ bands in GST was narrow, while the fwhm of those same bands in ICM, GCI, and GCIP were two to three times broader than those same corresponding bands in GST. The in-plane crystallite size ($L_a$) was inversely proportional to the $I_D/I_G$, and GST > GCI > ICM > GCIP were ranked large to small based on their $L_a$.

Figure 5, panel A showed the XRD spectra of GST and -400MSIP. The GST had an intense diagnostic graphite-002 reflection peak (G-002) at $\sim 26.5$°; its intensity was 5–33 times greater than that of G-004, G-100, or G-101 peaks. Thirty milligrams of GST had a stacking height of crystal (often called $L_c$) of 15.42 nm and an interlayer distance ($d$) of 0.334 nm. The Table 1. Physical and Morphological Characteristics of Graphite Standard (GST), and of -400 Mesh Spherical Fe Powder (-400MSIP)$^{a}$

|                  | Color, Lightness | Hardness, lb | Photo | SEM |
|------------------|------------------|--------------|-------|-----|
| Color calibrator | 97.2             | -            |       |     |
| Graphite Standard (GST, < 20 \(\mu\)m, synthetic, CAS #7782-42-5, Sigma-Aldrich) | 31.6 ± 0.1$^a$ | (n=3) |       |     |
| -400 mesh spherical Fe powder (-400MSIP, 99.99+ %, CAS # 7439-89-6, Sigma-Aldrich) | 44.1 ± 0.6$^c$ | (n=3) |       |     |
| Iron-carbon materials (ICM) produced during the CO$_2$ reduction step by our previous method (10 mg Fe powder and 525 °C for 6 h). | 39.7 ± 0.8$^b$ | 8.4 ± 0.9$^a$ | (n=15) | (n=10) |
| Graphite-coated Fe (GCI) produced during the CO$_2$ reduction step by our previous method (10 mg Fe powder and 525 °C for 6 h). | 30.8 ± 0.3$^a$ | 2.0 ± 0.4$^b$ | (n=15) | (n=10) |
| Graphite-coated Fe (GCIP) produced during the CO$_2$ reduction step by our optimized method$^{a}$ (5 g Fe powder and 500 °C for 3 h). | 31.2 ± 0.4$^a$ | - | (n=15) |     |

$^a$ Also shown are the physical and morphological characteristics of ICM and GCI by our previous method$^7$ and of GCIP by our optimized method.$^{13}$ The SEM pictures (2nd, -400MSIP; 4th, GCI; 5th, GCIP from top) were from ref 13.
Figure 3. FT-IR transmission spectra of GST, ICM, GCI, and GCIP. All spectra showed a water band at $\sim 1630$ cm$^{-1}$. All spectra, except that of ICM, showed a band at $\sim 1580$ cm$^{-1}$. This band corresponded to the sp$^2$ bond (C-C bond stretching) in graphite, and it demonstrated that GCI and GCIP each consisted of a carbon hexagonal structure.

$\sim 400$ MSIP, often called α-Fe or body-centered cubic lattice iron (bcc-Fe), had the α-Fe (110) reflection peak at $\sim 45^\circ$ and the α-Fe (200) reflection peak at $\sim 65^\circ$. Forty milligrams of α-Fe had a $L_c$ of 29.11 nm and an interlayer distance of 0.202 nm using α-Fe-110 as reference.

Figure 5, panel B summarized XRD spectra of the AMS targets ICM, GCI, and GCIP. Consistent with prior studies,$^{19–21}$ there was no evidence of a G-002 peak at $\sim 26.5^\circ$ in ICM, GCI, or GCIP, detecting only Fe$_3$C crystal with possible traces of Fe$_2$C. The α-Fe crystals within the range of $30^\circ$–$70^\circ$ (2θ).

Even if GCI spectrum lacked a G-002 peak, it still suggested the presence of g-C because FT-IR and Raman spectra of GCI showed a mix with some sp$^2$ and sp$^3$ carbon bond structures. As already seen from Figure 1, the i-C and g-C have an “amorphous (disordered)” and an “less ordered” carbon structure, respectively. The XRD spectrum of GCIP was very similar to those of ICM and GCI, and when the FT-IR and Raman spectra were considered together, the GCIP consisted of g-C and Fe$_3$C crystal. The G-101 reflection peak overlapped that of Fe$_3$C and of α-Fe (110), so it was difficult to individually differentiate graphite from α-Fe and Fe$_3$C at $\sim 45^\circ$.

Figure 5, panel C showed relative sensitivity toward 1:1, 1:10, and 10:1 ratios of GST/GCIP (including Fe$_3$C and α-Fe crystals) to Cu Kα, the X-ray source. A good quality XRD spectrum of graphite crystal (especially, G-002) required an AMS target that contained $\leq 2$ mg of graphite crystal. In all ratios of GST/GCIP, the G-002 reflection peak was much more sensitive than the reflection peaks of Fe$_3$C or α-Fe crystals. Finally, the graphite crystal mass of ICM, GCI, and GCIP was $\leq 2$ mg. The absence of a G-002 reflection peak indicated that ICM, GCI, and GCIP each consisted of a mix of Fe$_3$C crystal and g-C rather than of a graphite crystal. The ICM had more iron carbide (Fe$_3$C) crystal than g-C, and ICM produced low and variable ion current whereas the opposite was true for GCI and especially so for GCIP.

**DISCUSSION**

A variety of graphite-like substances are formed during the reduction of CO$_2$ to AMS targets depending on the conditions used. The types of graphite-like substances include filamentous,$^4$ fluffy, and firm or solid graphite;$^{22}$ these substances differ from one another in a variety of traits such as color, hardness, texture, SEM, FT-IR, Raman, and XRD spectra. To the extent that the traits affect the magnitude of the ion current produced, they are important so, we determined these traits in AMS targets produced using our previous$^7$ and optimized methods.$^{13}$ Our goal was to identify those traits in AMS targets that produced intense and reliable ion currents for accurate, precise, reliable, and high-throughput measurements of $^{14}$C using AMS.

Our previous method formed GCI (black, soft pellet, less uniform, fuzz) 80% of the time and ICM (gray, hard pellet, featureless carbon deposition) 20% of the time.$^7$ On the other hand, our optimized CO$_2$ reduction method formed only GCI (black, soft powder, uniform, fuzz) 100% of the time.$^{15}$ In general, the presence of a non-graphitic carbon or Fe$_3$C crystal resulted in the formation of a harder carbon structure than a compact graphitized carbon.$^{23,24}$

Graphite produced ion currents of 13.4 μA of $^{12}$C$^{1+}$/mg of C while a-C produced ion currents that ranged from 5.4 to 7.4 μA of $^{12}$C$^{1+}$/mg of C.$^3$ Furthermore, the presence of cobalt carbide in AMS targets produced still lower ion currents whose associated error in the $^{14}$C/$^{13}$C ratio was as high as 10%.$^5$ Because the ion current produced by ICM was 13% lower than that produced by

(19) Jaer, C.; Mutschke, H.; Huiskens, F.; Alexandrescu, R.; Morjan, I.; Dumitrache, F.; Barjega, R.; Soare, I.; David, B.; Schneeweiss, O. Appl. Phys., A: Mater. Sci. Process. 2006, 85, 53–62.
(20) Nikitenko, S. I.; Koltypin, Y.; Palchik, O.; Flerner, I.; Xu, X. N.; Gedanken, A. Angew. Chem., Int. Ed. 2001, 40, 4447–4449.
(21) Chung, U. C.; Kim, Y. H.; Lee, D. B.; Jeong, Y. U.; Chung, W. S.; Cho, Y. R.; Park, I. M. Bull. Korean Chem. Soc. 2005, 26, 103–106.
(22) Santos, G. M.; Mazon, M.; Southon, J. R.; Rifai, S.; Moore, R. Nucl. Instrum. Methods Phys. Res., Sect. B 2007, 259, 306–315.
(23) Franklin, R. E. Proc. R. Soc. London, Ser. A: Math. Phys. Sci. 1951, 209, 196–218.
(24) Wagner, D. B. Iron and Steel in Ancient China; E.J. Brill: London, 1993; pp 335–344.
GCI and by GCIP, isotopic fractionation occurred in ICM. In addition, the $F_m$ values of ICM (1.3355) were $\sim 0.4\%$ lower than those of Ox-2 SRM (1.3407), GCI (1.3429), and GCIP (1.3410), confirming prior reports.\textsuperscript{3,5} Furthermore, a large ratio of Fe/C ($15:1$) favored ICM formation,\textsuperscript{4,25} so by reducing the Fe/C ratio from $10:1$ to $5:1$, the formation of ICM was completely avoided.\textsuperscript{13} We did not find FT-IR, Raman, or XRD spectra of AMS targets in our search of the literature, so assumed that the ICM was mostly Fe$_3$C and some i-C or g-C rather than single graphite crystal. This observation prompted us to further investigate characteristics of AMS targets with FT-IR, Raman, and XRD.

**FT-IR and Raman Spectroscopy.** FT-IR enabled the vibration of aromatic structures in graphite to be investigated. For FT-IR, the KBr pellet showed a good spectrum by transmission measurements for the aromatic structure of graphite, because FT-IR spectra are not associated with crystal structure of graphite.\textsuperscript{26} The KBr pellet produced a band $\sim 1630$ cm$^{-1}$ (H$_2$O molecule) due to a moisture effect, and it was partially overlapped with the $\sim 1580$ cm$^{-1}$ of the graphite band.\textsuperscript{24} The FT-IR spectra of GCI and GCIP were consistent with that of GST in Figure 3 and with those in previous reports.\textsuperscript{26,27}

![Figure 3. Raman spectra of GST, ICM, GCI, and GCIP compared to published spectra of similar materials. Crystal size, in-plane, of graphite ($L_a$) = $C/(I_D/I_G)$, where $C \approx 4.4$ nm. Intensities of the $G'$ of commercially available glassy carbon are variable and are classified as narrow and strong or non-detectable.\textsuperscript{38,39} Soot is a highly disordered graphitic structure or amorphous carbon, because it consists of graphite-like crystalline and amorphous carbon domains. So, intensities of the D, G, G', D+G, and D'' bands from commercially available soot depend on the completeness solid carbon conversion to CO$_2$ and on the degree to which CO$_2$ is converted to graphite.\textsuperscript{16}](image3)

![Figure 4. Raman spectra from present study](image4)

| AMS target and carbon sample | Full width at half maximum for $D$, $G$, and $G'$ bands | Intensity | Crystal size | $L_a$ nm |
|-----------------------------|-------------------------------------------------------|-----------|--------------|---------|
| **Left panel** |
| GST                         | 43 21 ≈ 80                                            | 0.12      | 37.85        |
| ICM                         | 96 58 non-detectable                                  | 1.10      | 4.42         |
| GCI                         | 116 51 broad and weak                                 | 0.77      | 5.73         |
| GCIP                        | 98 66 broad and weak                                  | 1.01      | 4.40         |
| **Right panel** |
| Graphite powder\textsuperscript{38,39,41} | $\approx 60$ 15-30 70-90 0.06-0.2 $>$ 20          |           |              |
| Microcrystalline graphite\textsuperscript{38,39} | $\approx 60$ 35 90 0.4 $\leq 10$                   |           |              |
| Glassy carbon\textsuperscript{38,39} | $\leq 65$ 50-70 50 1.0-1.4 $= 4$                   |           |              |
| Disorder or amorphous carbon\textsuperscript{38,39} | $> 150$ 100 non-detectable - $< 2.5$                |           |              |

(25) Dee, M.; Ramsey, C. B. *Nucl. Instrum. Methods Phys. Res., Sect. B* 2000, 172, 449-453.

(26) Friedel, R. A.; Carlson, G. L. *J. Phys. Chem.* 1971, 75, 1149-1151.

(27) Smith, D. M.; Griffin, J. J.; Goldberg, E. D. *Anal. Chem.* 1975, 47, 233-238.

(28) Tuinstra, F.; Koenig, J. J. *Chem. Phys.* 1970, 53, 1126-1130.

(29) Gualberto, G. M.; Underhill, C.; Leung, S. Y.; Dresselhaus, G. *Phy. Rev. B* 1980, 21, 862-868.
∼1350 cm\(^{-1}\) and the other at ∼1580 cm\(^{-1}\). The band at ∼1350 cm\(^{-1}\) often combined with the band at ∼830 cm\(^{-1}\) to appear as a weak band at ∼2200 cm\(^{-1}\). Therefore, FT-IR spectra of graphite (devoid of a-C and of aromatic impurity) would be devoid of the 830-, ∼1350-, and ∼2200-cm\(^{-1}\) bands.\(^{26,27}\)

The Raman spectroscopy is more useful than FT-IR to determine the presence and features of graphite and disordered carbon, yet we found no information on iron carbide using the Raman in literature.\(^{30}\) The main features of graphitic materials in the Raman spectra were called G (∼1560 cm\(^{-1}\), \(E_{2g}\) mode, \(sp^2\)) and D (∼1360 cm\(^{-1}\), \(A_{1g}\) mode, \(sp^3\)) bands for visible excitation, while the a-C band (T band) appears at ∼1060 cm\(^{-1}\) seen in UV excitation.\(^{26,29,31,32}\) The iron carbide band was expected to appear between 200 and 600 cm\(^{-1}\) by Raman, but it was not detected because the low-energy vibration frequencies are too close to the laser line to be detected.\(^{33}\) The D band was first reported as a disorder-induced mode.\(^{28}\) The \(sp^3\) carbon bond content (D band) increased as amorphization and micro- and nanocrystalline graphitic materials were observed.\(^{34}\) The origin of the D band at 1350 cm\(^{-1}\) is not yet fully understood.\(^{35,36}\)

Figure 5. XRD spectra of GST, ∼400MSIP (reference standards, panel A), ICM, and GCI, using our previous method and GCIP using our optimized method\(^{13}\) (panel B), and 10:1 to 1:10 mixes of GST/GCIP (panel C). In panel A, GST had a G-002 reflection at ∼26.5° while ∼400MSIP had α-Fe-110 and α-Fe-200 reflections at ∼45° and ∼65°, respectively. Also, the G-002 reflection was more sensitive than either the α-Fe or Fe\(_3\)C crystal reflections. In panel B, ICM, GCI, and GCIP spectra had no (∼2 mg of graphite crystal) detectable G-002 reflection, but they did have detectable Fe\(_3\)C crystal and α-Fe crystal reflections at 35°–70°. Absence of a G-002 reflection indicated that ICM, GCI, and GCIP each consisted of a mix of Fe\(_3\)C crystal, a-C, and g-C rather than being a single graphite crystal. In panel C, the sum of GST+GCIP in the mix was 22 mg. In all ratios of GST/GCIP, the G-002 reflection was more sensitive than that of Fe\(_3\)C or α-Fe crystals. Also, the graphite crystal mass of ICM, GCI, and GCIP was ≤2 mg. The ICM was mostly Fe\(_3\)C crystal and the ICM contained less than half as much g-C compared to GCI and GCIP.

\(^{30}\) Park, E. Y.; Zhang, J. Q.; Thomson, S.; Ostrovski, O.; Howe, R. Metall. Mater. Trans. B 2001, 32B, 839–845.

\(^{31}\) Merhari, L.; Belorgeot, C.; Quintard, P. J. Mater. Sci. Lett. 1994, 13, 286–288.

\(^{32}\) Ferrari, A. C.; Robertson, J. Phys. Rev. B 2001, 64, 075414, 13 pages.

\(^{33}\) Bi, X. X.; Ganguly, B.; Huffman, G. P.; Huggins, F. E.; Endo, M.; Eklund, P. C. J. Mater. Res. 1993, 8, 1668–1674.
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statistically. However, the higher CO₂ reduction temperature to GCIP formed at 500 °C due to the treatment of graphite materials can reduce the intensity of the D band.

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Conclusions

AMS targets of GCI and GCIP each produced a mix of nanocrystalline graphite (g-C) and Fe₃C crystal based on FT-IR spectra, a clear presence of Fe₃C crystal in XRD spectra, we concluded that the ICM was mostly Fe₃C crystal with a minimal amount of sp² (g-C including nanocrystalline graphite) and sp³ (edge-carbon in graphite). The GCI and GCIP were also a mix of nanocrystalline graphite (g-C) and Fe₃C crystal based on FT-IR (C=C for 6 h compared a similar analysis with lower CO₂ reduction temperature and smaller mass ratio of C/Fe. Longer reduction time combined with higher CO₂ reduction temperature and smaller mass ratio of C/Fe may be useful to also minimize defects of in the graphite crystal.

As soon as AMS targets were produced, an overnight cooldown to room temperature (similar to annealing process) did not produce well-ordered larger graphite crystal compared to a 3-min cooldown. The graphitization yield and isotopic fragmentation were not influenced by the duration of a slow cooldown. Ion currents were affected more by the amount of graphite rather than its crystal size. Consequently a 3-min cooldown facilitated a high throughput for biological/biomedical applications of AMS. Previously a skilled analyst could prepare 60–150 AMS targets per day, while our optimized method produced as many as 270 AMS targets per d/skilled analyst.

CONCLUSIONS

AMS targets of GCI and GCIP each produced a mix of nanocrystalline graphite (g-C), and Fe₃C crystal, intense

(12) Kittel, C.; Klement, R. Phys. Rev. B 2000, 61, 14095–14107.

(35) Pimenta, M. A.; Dresselhaus, G.; Dresselhaus, M. S.; Cançado, L. G.; Jorio, A.; Salito, R. Phys. Chem. Chem. Phys. 2007, 9, 1276–1291.

(36) Hirlimann, C.; Jouanne, M.; Forrières, C. J. Raman Spectrosc. 1992, 23, 315–317.

(37) Reigh, S.; Thomsen, C. Phil. Trans. R. Soc., London A 2004, 362, 2271–2288.

(38) Escrigano, R.; Sloan, J. J.; Sidhique, N.; Sze, N.; Dudev, T. Vib. Spectrosc. 2001, 26, 179–186.

(39) Knight, D. S.; White, W. B., J. Mater. Sci. 1989, 4, 385–393.

(40) Nakamura, K.; Fujitsu, M.; Kitajima, M. Phys. Rev. B 1990, 41, 12261–12263.

(41) Nikiel, L.; Jagodziński, P. W. Carbon 1993, 31, 1313–1317.

(42) Santos, G. M.; Southon, J. R.; Griffin, S.; Beaupre, S. R.; Druffel, E. R. M. Nucl. Instrum. Methods Phys. Res., Sect. B 2007, 259, 293–302.
current consistently, with only traces of isotopic fragmentation, and accurately estimated the $F_m$ values. The GCIP produced by our optimized method$^{13}$ consistently produced $\sim$115 $\mu$A of $^{12}$C$^-$ and accurate and precise $F_m$ value with a throughput of up to 270 AMS targets per skilled analyst/d for biological/biomedical AMS applications.

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