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Early Neolithic wine of Georgia in the South Caucasus

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Chemical analyses of ancient organic compounds absorbed into the pottery fabrics from sites in Georgia in the South Caucasus region, dating to the early Neolithic period (ca. 6,000-5,000 BC), provide the earliest biomolecular archaeological evidence for grape wine and viniculture from the Near East, at ca. 6,000-5,800 BC. The chemical findings are corroborated by climatic and environmental reconstruction, together with archaeobotanical evidence, including grape pollen, starch, and epidermal remains associated with a jar of similar type and date. The very large-capacity jars, some of the earliest pottery made in the Near East, probably served as combination fermentation, aging, and serving vessels. They are the most numerous pottery type at many sites comprising the so-called "Shulaveri-Shomutepe Culture" of the Neolithic period, which extends into western Azerbaijan and northern Armenia. The discovery of early sixth millennium BC grape wine in this region is crucial to the later history of wine in Europe and the rest of the world.

Neolithic | wine | viticulture | Georgia | Near East

Following the last Ice Age, the Neolithic period in the Near East (ca. 10,000–4,500 BC) was a hotbed of experimentation, especially in the mountainous region extending west to east from the Taurus Mountains of southeastern Anatolia through the South Caucasus and northern Mesopotamia to the Zagros Mountains of northwestern Iran (e.g., refs. 1 and 2, including pertinent references). As the climate moderated and precipitation levels increased, especially between ca. 6,200–4,200 BC (*SI Appendix*), humans established year-round settlements. Permanent habitation allowed for a host of recently domesticated plants—including the "founder crops" of barley, einkorn wheat, emmer wheat, chickpea, pea, lentil, flax, and bitter vetch—to be efficiently raised, harvested, and stored. These developments were crucial in jump-starting the millennia-long upheaval and changes in human subsistence and culture known as the "Neolithic revolution" (3, 4).

Sedentary life, made possible by new, assured plant resources, was also accompanied by advances in the arts and crafts, such as architecture, weaving, dyeing, stone working, and woodworking. The invention of fired clay (pottery) containers sometime during the early seventh millennium BC (5, 6) had profound implications for processing, serving, and storing food and drink.

Human exploitation and cultivation of plants was not confined to staple cereals and legumes during the Neolithic. Fruits, nuts, tubers, herbs, and tree products are well-attested at Neolithic sites throughout the larger region. Among the fruit species, the wild Eurasian grape (*Vitis vinifera* sp. *sylvestris*) stands out, because its domestication as *V. vinifera* sp. *vinifera* became the basis of a widespread "wine culture" throughout the Near East and Egypt (1), which later spread to east Asia and across the Mediterranean to Europe (7–9), and then later to the New World. Today, there are some 8,000–10,000 domesticated cultivars of wine, raisin, and table grapes, with a range of colors from black to red to white. These cultivars owe their origins to

human selection and accidental crosses or introgression between the incoming domesticated vine and native wild vines. These varieties account for 99.9% of the world's wine production and include famous Western European cultivars such as Cabernet Sauvignon, Sangiovese, Tempranillo, and Chardonnay (10).

The Near Eastern uplands have been described as the "world center" of the Eurasian grape (11), based on where the wild plant thrived and achieved its greatest genetic diversity. Indeed, DNA studies have shown that the wild vine of Anatolia is genetically closer to Western European cultivars than its wild counterpart there (12–16). Many cultivars in Georgia also have a close relationship to those in the West, including Pinot Noir, Nebbiolo, Syrah, and Chasselas (12).

Two important questions remain to be answered. Can more narrowly defined mountainous areas of greater Mesopotamia and the Fertile Crescent be delimited where the Eurasian grape first began to be made into wine and where it was subsequently domesticated? If so, when did these developments occur?

Archaeological Samples Chosen for Analysis

Our investigation, part of a larger Georgian project (17), sought to answer these questions by focusing on two archaeological sites

Significance

The earliest biomolecular archaeological and archaeobotanical evidence for grape wine and viniculture from the Near East, ca. 6,000–5,800 BC during the early Neolithic Period, was obtained by applying state-of-the-art archaeological, archaeobotanical, climatic, and chemical methods to newly excavated materials from two sites in Georgia in the South Caucasus. Wine is central to civilization as we know it in the West. As a medicine, social lubricant, mind-altering substance, and highly valued commodity, wine became the focus of religious cults, pharmacopoeias, cuisines, economies, and society in the ancient Near East. This wine culture subsequently spread around the globe. Viniculture illustrates human ingenuity in developing horticultural and winemaking techniques, such as domestication, propagation, selection of desirable traits, wine presses, suitable containers and closures, and so on.

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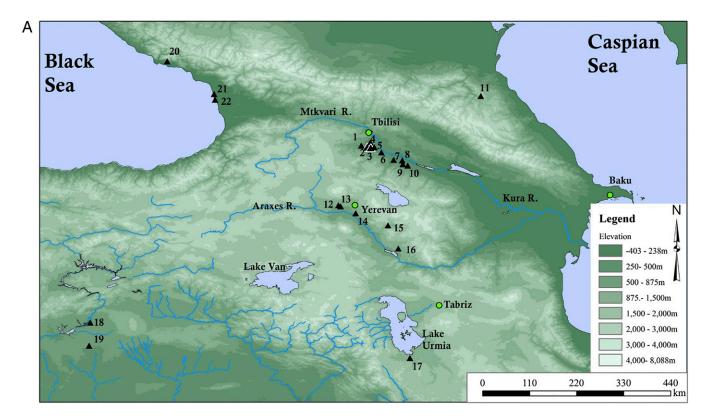
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that were occupied during the earliest Pottery Neolithic period in Georgia, the so-called "Shulaveri-Shomutepe Culture" (SSC), dated to ca. 5,900–5,000 BC (18–20). The two sites are Shulaveris Gora, which gives its name to the period together with Shomutepe approximately 50 km downstream on the Kura River, and Gadachrili Gora (21). These sites are located within 2 km of one another in the province of Kvemo (Lower) Kartli, roughly 50 km south of the modern capital of Tbilisi (Fig. 1).

Each is a small village, approximately 1 ha in area, of closely spaced mudbrick circular structures, 1–5 m in diameter, with interspersed pits and courtyards. The buildings are believed to be domestic residences, and the pits assumed to be for storage and/ or refuse.

Fertile, rolling hills surround the sites on a high plateau at an altitude of >1,000 m ASL. Gadachrili Gora is presently bifurcated by the Shulaveris Ghele, a seasonal tributary of the



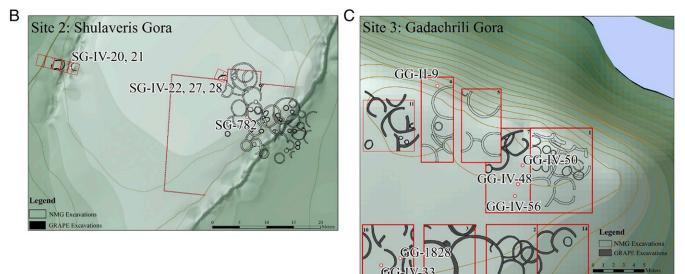


Fig. 1. Map of Shulaveri-Shomutepe Culture sites and other sites mentioned in the text (*A*) and the early Neolithic settlements of Shulaveris Gora (*B*) and Gadachrili Gora (*C*) showing the locations of the analyzed jar sherd samples that were positive for tartaric acid/tartrate. Site names: Arukhlo (1), Shulaveris Gora (2), Gadachrili Gora (3), Dangreuli Gora (4), Imeris Gora (5), Khramis Didi-Gora (6), Shomutepe (7), Haci Elamxali Tepe (8), Göytepe (9), Mentesh Tepe (10), Chokh (11), Aratashen (12), Aknashen (13), Masis Blur (14), Areni-1 (15), Kül Tepe (16), Hajji Firuz Tepe (17), Nevali Çori (18), Göbekli Tepe (19), Gudau River (20), Pichori (21), and Anaklia (22). GRAPE, Gadachrili Gora Regional Archaeological Project Expedition; NMG, National Museum of Georgia; R, river. Red lines indicate excavated areas and squares.

Khrami River that runs into the Kura, while Shulaveris Gora is roughly 0.5 km from the stream. The climate today is semiarid (steppe), with an annual rainfall of 350–550 mm and an average temperature of approximately 13 °C. Milder, better-watered conditions prevailed during the period ca. 5,900–5,000 BC (*SI Appendix*). The Eurasian grapevine was well adapted to the ancient climate and remains well adapted to the modern climate.

As is our standard practice in biomolecular archaeological investigations (22), we strove to obtain the best-dated, best-provenienced, and best-preserved samples possible. These criteria were met to a varying extent in this study. For example, we had previously analyzed two sherds (SG-16a and SG-782; Fig. 2 B-C and Table 1) from the 1960s excavations at Shulaveris Gora, which we designated as "borderline positives" for tartaric acid/tartrate (1), the principal biomarker of grape/wine in the Near East (*SI Appendix*), because of conflicting results from the less-sensitive chemical techniques that we used at that time. Moreover, the customary practice at that time was to "clean" sherds by washing them in dilute hydrochloric acid to remove calcium carbonate and other postburial accretions. In the process, ancient organics might well have been altered, even destroyed, to

give "false positives." It was also later learned that the sherd with the highest apparent level of tartaric acid/tartrate (SG-16a) was collected from the surface of the site. Besides compromising the dating of this sherd, this also called into question the extent to which it had been subjected to environmental contamination and exposure to rain, which might have caused increased microbial activity and an elevated tartaric acid/tartrate content.

The opportunity to learn more and put the biomolecular archaeological investigation on a firmer, multidisciplinary foundation came when excavations at Gadachrili and Shulaveri were renewed in 2012–2013 and 2015–2016 (17). Many more radiocarbon dates from well-defined occupational contexts were obtained; coupled with advances in calibration curves and statistical evaluation, this has allowed for construction of a much tighter chronology for the early Neolithic than had been proposed in earlier publications (*SI Appendix*). Excavation and archaeobotanical techniques have also advanced since the 1960s, providing a finer-grained picture of how artifacts and ecofacts (i.e., plant and animal remains) were deposited and subjected to geological and chemical processes, as well as to human activity.



Fig. 2. (A) Representative early Neolithic jar from Khramis Didi-Gora (field no. XXI-60, building no. 63; depth, -5.45 to -6.25 m). (B) Jar base SG-16a, interior and cross-section. (C) Jar base SG-782, exterior. Note the textile impression on the base. (D) Jar base GG-IV-50, interior. (Photographs by Mindia Jalabadze and courtesy of the National Museum of Georgia.)

Table 1. Georgian early Neolithic pottery positive for tartaric acid by LC-MS-MS and their associated soil samples

| Sample no. | • | | - | | | | • | |
|---------------------------|-----------------|------------------------|-----------------|------------------------|-----------------------------------|--------------------------------|-----------------------------------|---------------------------------|
| | Date (BC) | Provenience | Pottery type | Extract weight (mg) | Tartaric acid (ng/mg residue)* | Malic acid (ng/mg residue)* | Succinic acid (ng/mg residue)* | Citric acid (ng/mg residue)* |
| | | | | | | | | |
| GG-II-9, body sherd | ca. 5900-5750 | Square BB-27, -2.73 m | Jar base sherd | NA | 134 ± 11* | 715 ± 86* | 596 ±25* | 182 ± 3* |
| GG-II-9, soil | ca. 5900-5750 | Square BB-27, -2.73 m | Associated soil | NA | $20 \pm 2*$ | 491 ± 7* | 630 ± 21* | $10 \pm 1*$ |
| GG-IV-33, disk base sherd | ca. 5700-5500 | Square 10, Locus 4 | Jar base sherd | 1.2 | 87 ± 6 | 998 <u>+</u> 47 | 165 ± 13 | 186 ± 6 |
| GG-IV-62, soil | ca. 5700-5500 | Square 10, Locus 4 | Associated soil | 0.8 | 7 ± 1 | 193 ± 33 | 32 ± 5 | 9 ± 0 |
| GG-IV-50, pedestal base | ca. 5700-5500 | Square 7, Locus 2 | Jar base sherd | 1.2 | 17 ± 1 | 170 <u>+</u> 13 | 31 ± 4 | 45 ± 1 |
| GG-IV-51, soil | ca. 5700-5500 | Square 7, Locus 2 | Associated soil | 4.6 | 5 ± 0 | 91 ± 7 | 16 ± 0 | 5 ± 1 |
| GG-IV-48, pedestal base | ca. 5700-5500 | Square 7, Locus 2 | Jar base sherd | 4.3 | 4 ± 1 | 50 <u>+</u> 1 | 22 <u>+</u> 1 | 6 <u>+</u> 1 |
| GG-IV-54, soil | ca. 5700-5500 | Square 7, Locus 2 | Associated soil | 4.5 | 1 ± 0 | 23 ± 1 | 7 ± 1 | 1 ± 0 |
| GG-IV-56, flat base | ca. 5700-5500 | Square 7, Locus 1 | Jar base sherd | 6.3 | 39 ± 0 | 369 <u>+</u> 22 | 54 ± 0 | 51 <u>+</u> 0 |
| GG-IV-46, soil | ca. 5700-5500 | Square 7, Locus 1 | Associated soil | 2.2 | 19 ± 0 | 312 ± 16 | 34 ± 4 | 20 ± 0 |
| Shulaveris-Gora | | | | | | | | |
| SG-16a, flat base | Early Neolithic | Surface | Jar body sherd | NA | 55 ± 1 [†] | 2028 ± 71 ⁺ | 198 \pm 4 ⁺ | 58 ± 1 [†] |
| SG-782, pedestal base | ca. 5900-5750 | Square BB, –0.8 m | Jar body sherd | NA | $8 \pm 0^{\dagger}$ | $387 \pm 14^{\dagger}$ | 56 \pm 4 [†] | 15 ± 0 ⁺ |
| SG-IV-20, body sherd | ca. 5900-5750 | Square 2, Locus 2 | Jar base sherd | 6.1 | 4 ± 0 | 97 ± 2 | 12 ± 1 | 34 ± 0 |
| SG-IV-21, soil | ca. 5700-5500 | Square 2, Locus 2 | Associated soil | 7.1 | 3 ± 0 | 56 ± 0 | 12 ± 1 | 5 ± 1 |
| SG-IV-22, soil | ca. 5700-5500 | Soil, Neolithic levels | Site soil | 9.6 | 2 ± 0 | 17 ± 2 | 3 ± 0 | 1 ± 0 |
| SG-IV-27, soil | ca. 5700-5500 | Soil, Neolithic levels | Site soil | 8.8 | 2 ± 0 | 18 ± 1 | 4 ± 0 | 1 ± 0 |
| SG-IV-28, soil | ca. 5700-5500 | Soil, Neolithic levels | Site soil | 14.6 | 1 ± 1 | 9 ± 1 | 4 ± 1 | 1 ± 0 |

Numbers in bold highlight concentrations for ancient sherds that are higher than their corresponding soils. NA, not applicable.

*Except for the GG-II-9 samples, which are reported as nanograms of organic acid per gram of sherd/soil material (ng/g or ppb), all concentrations are cited as ng/mg (ppm) of extracted residue.

[†]Sherds were extracted in toto.

Pottery is the essential starting point of many biomolecular archaeological investigations. Barring the recovery of discernible physical residues of natural products constituting a food or drink, pottery has the advantage of being porous and an ionic (zeolitelike) material that absorbs liquids in particular and preserves them from environmental contamination for millennia until they are chemically extracted (see below).

Pottery had some additional advantages for our study. The plasticity of the clay is ideal for producing vessel shapes suited to specific purposes, and once fired, the material is virtually indestructible. The beginning stages of pottery making in the Near East are attested at Gadachrili and Shulaveri. The pottery is well-made and functional, implying that it derives from even earlier industrial developments, possibly from a nearby mountainous region of Turkey, Mesopotamia, or Iran. Although the vessels were handmade, textile impressions on the bottoms of some bases indicated that they were probably turned on a slow wheel.

Fortunately, it has been possible to reconstruct in its entirety what is likely the principal jar type of the period. Large jars, like the one from Khramis Didi-Gora shown in Fig. 24, are among the most common shapes in the pottery corpora of Gadachrili, Shulaveri, and other SSC sites. They can be very large; for example, the Khramis Didi-Gora specimen is nearly 1 m tall and 1 m wide, with a volume exceeding 300 L. Strangely, their bases, which are flattened or low disks or low pedestals, can be relatively small and seemingly unstable; the diameter of the Khramis Didi-Gora jar base is only one-quarter of its overall diameter at its widest point (*Discussion and Conclusions*).

Globules and strips of clay were sometimes applied as plastic decorations to the exterior surfaces of jars, especially very large ones. Fig. 2A shows 10–15 clay globules enclosed within semicircular strips at intervals around the mouth of the vessel. This motif has been interpreted as a schematic grape cluster. Small central indentations of individual globules on other jars might then represent the attachment points of bunches of berries to their pedicels. The larger knobs in the intervening spaces could indicate how a cover or lid made of an organic material (perhaps leather or cloth) was held down. Another jar from Khramis Didi-Gora is thus far unique in showing a stick-like figure with upraised arms beneath vertical lines of globules (*SI Appendix*, Fig. S1). Could this be a Neolithic rendition of a popular motif, seen on modern monuments and buildings throughout Georgia today, in which jubilant, dancing figures are seen cavorting under trellised grapevines? Chemical analysis was clearly needed as a check on any fanciful interpretations.

The pottery fabrics of all vessel types, including bowls and a range of different-sized jars with both narrow and wide mouths, are moderately well-fired, occasionally straw-tempered, and rarely polished (burnished) on their reddish-yellowish exteriors (Fig. 2 B–D). Interiors are generally blackish-grayish due to the narrow mouths of jars cutting off oxygen, and variously sized reduction splotches of the same colors on the exterior surfaces pointed to open-firing rather than kiln-firing (23). Interior reddish residues on the lower halves and bases of jar interiors were infrequently observed, but were suggestive of precipitates from liquid contents.

As might be anticipated, pottery was not produced on a large scale in the early Neolithic, and relatively little pottery has been recovered from these sites compared with later eras. Unlike undisturbed burials with intact vessels, human occupation, especially when a site has been intensely inhabited over centuries, usually results in whole vessels being broken into sherds and dispersed.

Based on considerations of good context and preservation, assured dating, special features such as decoration, and availability, 18 jars (6 body sherds and 12 base sherds) were sampled from the 2012–13 and 2014–2016 seasons at Gadachrili, along with one jar base sherd from the more limited 2016 season at Shulaveri. Bases were most desirable because materials settling out from a liquid were most likely to have accumulated on their interiors. Body sherds were less definitive, since they might come from the lower or upper part of a vessel. The sherds, which were not washed in the field, were accompanied by soil samples, collected from the same contexts but separated from the sherds, so as to provide a check on possible environmental contamination and background organic acid production by microorganisms.

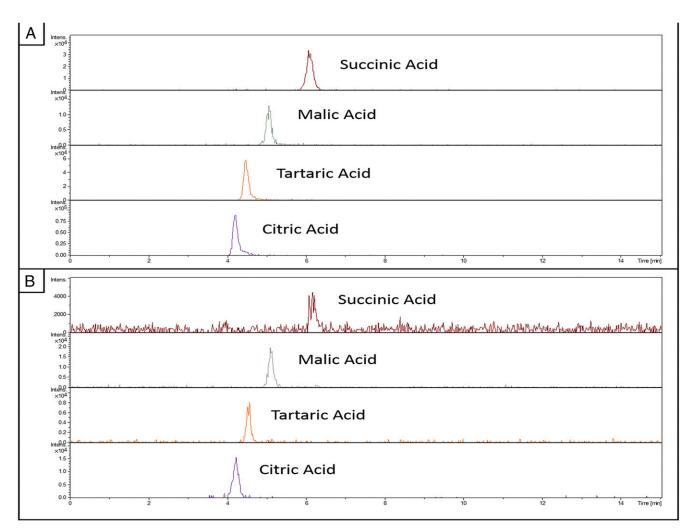


Fig. 3. Extracted ion chromatograms (\pm 0.005-Da window) for 5 μ M standard solutions (A), using the theoretical mass of deprotonated tartaric, malic, succinic, and citric acid, compared with jar sherd sample GG-IV-50 (B). All four organic acids were positively detected and quantified in this sample. Intens, intensity.

The two putative "positive" samples from the 1960s excavations at Shulaveri (one base sherd and one body sherd) were included in our analytical corpus for reanalysis with our stricter protocols and more sensitive instrumentation. Three general soil samples from Neolithic levels served as controls. Soils at both Shulaveri and Gadachrili were of the gray cinnamonic dark type.

Relative Chronology and Absolute Dating

Given our claim to have identified the earliest grape wine in the Near East (ca. 6,000–5,800 BC), it is crucial to put our findings on a solid chronological footing. Our primary reliance on short-lived botanical samples, well-defined archaeological contexts, and a Bayesian analysis of the composite data ensure that all of the analyzed samples from Shulaveris Gora and Gadachrili Gora belong to the first half of the sixth millennium BC.

Kiguradze (18) first developed a five-phase chronological model for the SSC based on the Kvemo Kartli group of sites in the Kvemo (Lower) Kartli province: Shulaveris Gora, Imeris Gora, and Khramis Didi Gora. His chronology of the relative phasing of the sites was anchored by 10 radiocarbon dates, which were carried out in the early days of the technique's development by Soviet laboratories (24). Renewed excavations at Gadachrili Gora in the same region provided an additional three calibrated dates to the corpus (21), and the 2016 excavation of Gadachrili and Shulaveris Gora added another nine calibrated dates (Datasets S2 and S3).

Even though different laboratories carried out the 22 analyses with different levels of precision and calibration, most of the dates approximated Kiguradze's original phasing and dating. A Bayesian analysis (25) of the determinations enabled Kiguradze's dates to be recalibrated with the most recent 2016 dates using OxCal v. 4. 3.2 and IntCal 13 (26–28), as shown in composite *SI Appendix*, Fig. S10 and Dataset S2. This analysis suggests that Kiguradze's fivefold model should be expanded to six phases, including an earlier phase 1 extending back into the seventh millennium BC, which is consistent with radiocarbon dates from Azerbaijan (29). Phase 1's upper limit remains to be defined by additional radiocarbon determinations.

Chemical Results

After sample extraction, ancient organic compounds were identified by a combination of chemical techniques, including Fouriertransform infrared spectrometry (FT-IR), gas chromatography-mass spectrometry (GC-MS), and liquid chromatography linear ion trap/ orbitrap mass spectrometry (LC-MS-MS) (*SI Appendix*).

Our previous FT-IR results for base sherds SG-16a and SG-782 from the excavations at Shulaveris Gora in the 1960s had been promising for the presence of tartaric acid/tartrate. In 2016,

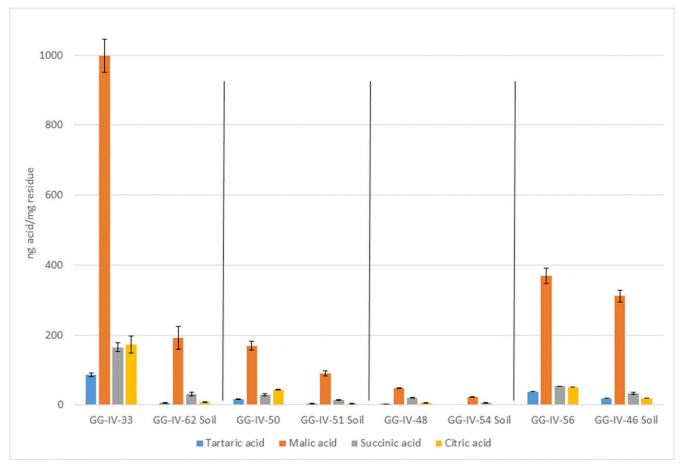


Fig. 4. Organic acid distribution for the LC-MS-MS-analyzed ancient jar base samples that were positive for tartaric acid/tartrate at Gadachrili Gora, compared with their associated soil samples. Concentrations are reported as nanograms of organic acid per milligram of extracted residue from sherd/soil material, and errors as the SD of two measurements. Note that the GG-II-9 samples (Table 1) are omitted from this graphical representation, because their data were reported as nanograms of organic acid per gram of extracted sherd/soil material.

we reran the samples, together with Neolithic soil samples from the site collected during the 2016 season. As shown in *SI Appendix*, Fig. S2, the spectrum of SG-782 had more pronounced straight-chain carbon-hydrogen stretch bond peaks at 2,920 and 2,850 cm⁻¹ compared with soil, an indication that the extracted ancient sample is relatively richer in hydrocarbons. The characteristic tartaric acid doublet-carbonyl stretch bond peaks at 1,716 and 1,734 cm⁻¹ were apparent for the ancient sherd, as was the hydroxyl bend at 1,452 cm⁻¹. Tartrate was identified by the carbonyl stretch bond peaks at 1,636 and 1,598 cm⁻¹, as well as the carboxylate stretch at 1,380 cm⁻¹. In contrast, the soil spectrum had very ill-defined absorptions in these regions, which might be variously interpreted.

Comparable spectra were observed for the Gadachrili sherds (e.g., Fig. 2D) that were positive for tartaric acid by LC-MS-MS (Table 1).

Searches of our FT-IR databases also yielded excellent statistical "matches" of the ancient spectra from both sites to those of other ancient and modern wine samples and synthetic tartaric acid and tartrate (*SI Appendix*).

Our recent GC-MS analyses were uninformative about the original contents of the jars from both sites. Fatty acids predominated in all of the samples, especially palmitic and stearic acids. The chromatogram (*SI Appendix*, Fig. S3) of jar base GG-IV-50, which was positive for tartaric acid by LC-MS-MS, is representative. Branched and unsaturated fatty acids also might occur, together with the occasional alcohol, high-numbered hydrocarbon, hopane-related triterpenoid (generic to plant cell walls), C_9 and C_{10} dioic acids (breakdown products of oleic acid), and nonspecific stigmasterol (a plant steroid). Contaminants, such as phthalate (a plasticizer ingredient of the bags in which the sherds were stored) and behenic acid (used in hand moisturizers), were ever-present.

A comparison of the chromatogram of the ancient sherd (*SI Appendix*, Fig. S4) with that of its associated soil sample (GG-IV-51) shows that the soil is richer in organics, especially high-numbered hydrocarbons (C_{27} – C_{33}) at retention times exceeding 20 min. The soil compounds are likely of modern origin. Fatty acids and n-alkanes occur widely in plants and animals, and are produced by microorganisms; they are not definitive for a grape-derived product.

The LC-MS-MS analyses proved to be most productive. Altogether, five base sherds from Gadachrili and three from Shulaveri were shown to be positive for tartaric acid and other organic acids (malic, succinic, and citric acid) found in grape/wine.

The presence of the four acids in the ancient samples is demonstrated by the exact correspondence of retention times for their extracted ion chromatograms with those of modern standards (Fig. 3). As seen in Fig. 4 and Table 1, the tartaric acid content of the positive sherds from Gadachrili (GG-II-9, GG-IV-33, GG-IV-48, GG-IV-50, and GG-IV-56) exceeded that of their corresponding background soil samples by 3.4- to 12.4-fold. At Shulaveri (Fig. 5), the tartaric acid level of SG-16a was 44 times that of the average of three Neolithic soil samples (SG-22,

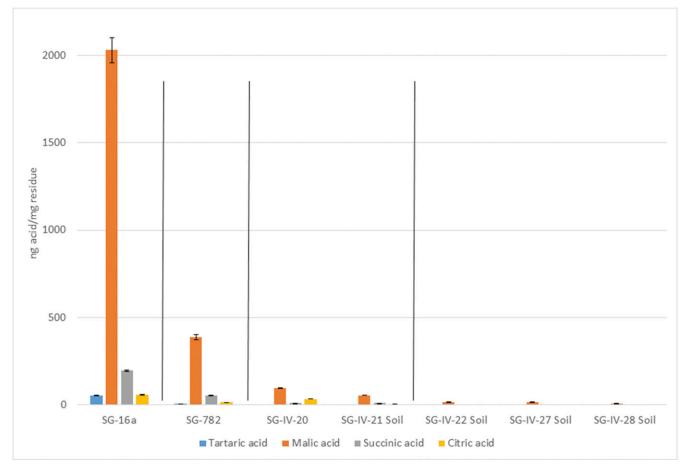


Fig. 5. Organic acid distribution for the LC-MS-MS-analyzed ancient jar base samples that were positive for tartaric acid/tartrate at Shulaveris Gora, compared with their associated soil samples. Concentrations are reported as nanograms of organic acid per milligram of extracted residue from sherd/soil material, and errors as the SD of two measurements.

SG-27, and SG-28). In contrast, the tartaric acid content of SG-IV-20 was only 1¹/₃ times that of its associated soil (SG-IV-21) and very low (4 ng/mg residue). Any variability in microbial soil activity (*SI Appendix*) might well lead to SG-IV-20 being classified as negative.

Negative results (not shown here) were also obtained, including 11 Gadachrili samples (five jar bases and six body sherds) with tartaric acid concentrations below those of their associated soil samples. Two other bases from this site, GG-IV-49 and GG-IV-60, did not contain any detectable levels of tartaric or the other organic acids.

Two of the bases from Shulaveris Gora (SG-16a and SG-782) were extracted as complete sherds (in toto), as was our customary procedure in the late 1990s, and were then analyzed by high-resolution LC-Orbitrap MS-MS (Table 1). The Shulaveri soils were markedly lower in abundance of the four organic acids than the soils at Gadachrili. Rainy conditions at the time of collection appear to have contributed to this difference (*SI Appendix*). High levels of tartaric acid, especially for SG-16a, provide very strong evidence for the presence of ancient grape/ wine in this jar and others from Gadachrili (e.g., GG-IV-33).

Archaeobotanical Results

If grapes were exploited to make wine or used as a food source at Shulaveris Gora and Gadachrili Gora, as well as other SSC sites, then corroborative archaeobotanical evidence—seeds (pips), grapevine wood, even desiccated remains, such as skins—might be expected. Thus far, no grape pips, which have been confirmed to be Neolithic by radiocarbon dating, have been recovered from an SSC site. Those that have been excavated, including both uncarbonized and carbonized specimens, have been shown to be post-AD 1600, or "modern" in date (*SI Appendix* and Dataset S3). Only two later Middle Bronze pips were in accordance with their archaeological dating, one an uncarbonized seed with wild features per geometric morphometric analyses (ref. 24 and *SI Appendix*) from the site of Dicha Gudzuba in the port city of Anaklia and the other a carbonized pip from Pichori, north of Anaklia on the Black Sea Coast (Fig. 1), which has not yet been analyzed by geometric morphometry but appears to be of the domesticated type.

To date, the recovery of single carbonized grape pips appears to be the rule at SSC sites, including Mentesh Tepe (wild morphology; ref. 30), Göytepe (uncertain morphology; ref. 29), and Haci Elamxanli Tepe (uncertain morphology; ref. 29) in Azerbaijan. Only Aratashen in Armenia, with two pips (wild), has yielded more than one (31). Carbonized grape wood at Mentesh Tepe (30) points to grapevines growing at the site or in its environs. None of these specimens has been radiocarbon-dated, however. Possible explanations for the relative lack of grape seeds in the early Neolithic, especially given the prevalence of well-dated cereal grains from the period, are addressed below.

The archaeobotanical database for grapes at SSC sites was expanded to include evidence of pollen, starches, and phytoliths by analyzing soils and artifacts from the 2016 Gadachrili and Shulaveri excavations (*SI Appendix*). These data provide direct, contemporaneous evidence that grapes—whether wild or domesticated is not yet clear—were an important natural resource at these sites.

Grape pollen (*SI Appendix*, Fig. S7 *A* and *C*) is widespread and abundant in many of the excavated early Neolithic contexts at both sites (e.g., locus 9 at Shulaveri; *SI Appendix*, Fig. S8*A*), but is absent from the modern top soils of the sites (*SI Appendix*, Fig. S9). The nearest grapevines in the area today are several kilometers away, and it has been demonstrated that grape pollen is distributed by wind over a short distance (32, 33). It can be concluded that the pollen from the Neolithic level is ancient. Moreover, agglomerations of pollen (*SI Appendix*, Fig. S7*A*), which are best interpreted as the remains of grape flowers, imply that grapes were growing near or even at the sites in the Neolithic. Supporting evidence for these conclusions is provided by results that are consistent with grape starch (*SI Appendix*, Fig. S7*B*) and grapevine epidermis (*SI Appendix*, Fig. S7*D*).

Pollen, palynomorphs, and nonpollen microfossils were also extracted by standard palynological analysis combined with acetolysis (*SI Appendix*) from a jar body sherd (serial no. 1828) at Gadachrili. It was excavated from a sealed context (square 10, locus 7, lot 22) inside a circular Neolithic building. Its spectrum of tree, cereal, and herbaceous pollen (*SI Appendix*, Fig. S8*B*) is similar to that of a stone grinder fragment from nearby squares 2 and 3 (locus 35). Unlike the jar sherd, however, the grinder did not yield any grape starch, grapevine epidermis, or remains of fruit flies (*Drosophila melanogaster*) (*SI Appendix*, Fig. S7*E*), which are attracted to sugar and alcohol. It can be hypothesized that the jar once contained grape wine and/or beer (compare ref. 34). Grape juice readily ferments into wine (*SI Appendix*).

Based on this microbotanical evidence, two reasonable, parsimonious inferences can be made: that grapevines were growing close to the Georgian sites, possibly inside the villages, and that their fruit was used as a food source. Combined with the chemical evidence for a grape product inside several jars, which would have served well as liquid containers, grape wine was likely one of the intended products, especially in light of the "wine culture" that emerged later in this area and throughout the Near East and Egypt.

Discussion and Conclusions

Previously, the earliest evidence for grape wine in the Near East was from the early Neolithic village of Hajji Firuz Tepe in the northwestern Zagros Mountains of Iran, ca. 5,400-5,000 BC (1, 35). Six jars, two of which were analyzed and showed the presence of tartaric acid/tartrate and a tree resin, had been embedded in the earthen floor along one wall of a "kitchen" of a Neolithic mudbrick house. Each jar when full had a volume of approximately 9 L-altogether, approximately 55 L for an average household. If that amount of wine is multiplied many times over by the houses throughout the settlement, then the production level would have already been relatively large scale at this early date. Either wild grapes were plentiful in the area or the Eurasian grapevine was already being intentionally cultivated or even domesticated. Hajji Firuz lies within the ancient and modern distribution zone of the wild grape, as established by pollen cores from nearby Lake Urmia.

The Hajji Firuz jar shapes are also well suited for vinification and wine storage, implying that they are part of an earlier industrial tradition. Their narrow, high mouths could have been stoppered with clay (some possible examples with the same diameter as the mouths of the jars were found nearby) or covered.

Hajji Firuz is only approximately 500 km from Shulaveri and Gadachrili, and even closer to sites in Armenia and Azerbaijan. These sites also lie within the zone of the wild grape, as does the mountainous region of northern Mesopotamia and, farther afield, the Taurus Mountains of eastern Anatolia. Now that wine jars from as early as ca. 6,000 BC have been confirmed for Gadachrili and Shulaveri, preceding the Hajji Firuz jars by half a millennium, the question might be asked which region has priority in the dis-

covery and dissemination of the "wine culture" and the domesticated grape. It is impossible to assign priority to any of these regions at this stage in the investigation; much more excavation and the collection of wild grapevines for DNA analysis are needed.

One disparity between the analyses of Hajji Firuz and Georgian jars is that the latter showed no signs of a tree resin or any other additive, according to the GC-MS analyses. Pine and terebinth saps were commonly added to wine throughout antiquity. They acted as antioxidants to keep the wine from going to vinegar, or barring that, to cover up offensive aromas and tastes. The tradition continues today only in Greece as retsina.

The Hajji Firuz jars were found partly buried in an earthen floor. No evidence has yet been found of how the Shulaveri and Gadachrili jars were positioned or whether they were partly or fully buried underground, as is the common practice for making so-called *qvevri* ("large jar") wine today in Georgia. The very small, flat bases of the ancient jars, often disks or low pedestals, seem inadequate to independently support a vessel full of liquid, so a case could be made for burying them. But then why even provide them with such unstable bases, unless these were decorative like the plastic decorations on some examples?

The earliest archaeological evidence for *qvevri* winemaking in Georgia is Iron Age in date, specifically the eighth to seventh centuries BC By Roman and Byzantine times, *qvevris* had become very popular throughout the Near Eastern and Mediterranean worlds; for example, excellent examples have been unearthed at Pompeii. Strangely, however, no examples of large jars buried underground like those at Areni in Armenia have been found in Georgia for the 5,000-y period from the Neolithic period to Iron Age times.

Based on ancient Egyptian frescoes, the earliest pictorial record of winemaking in the world, fermenting wine in mediumsized jars (amphoras) totally above ground was the preferred method since ca. 3,000 BC (1, 36). Given that Canaanites introduced viticulture, winemaking, and the amphora ("Canaanite jar") to Egypt, it can be assumed that they performed vinification and storage of wine, as the Phoenicians did later, in the same way.

The breakthrough came when numerous underground jars were found inside caves at Areni in a mountainous region of Armenia (37). Desiccated (uncarbonized) grapevine wood, dating to ca. 4,000 BC, together with pips and chemical evidence by LC-MS-MS of tartaric acid/tartrate and the red pigment malvidin, left no doubt that we now had partial evidence for the previously "empty" transitional period. The technology was ingenious: humans had laid out plaster floors for pressing the grapes and running the unfiltered juice into underground jars. Whether similar evidence will eventually be found in Georgia and Azerbaijan, elsewhere in the SSC area, or in the extended mountainous region remains to be seen.

The prominence of cereals in the early Neolithic SSC sites was likely due to a combination of factors. Barley and the wheats (einkorn and emmer) were domesticated very early in the Near East, perhaps by ca. 10,000 BC. They provided the all-important ingredients for beer and bread, staples that were produced in quantity in succeeding periods. The probable later domestication of the grapevine, combined with the fact that it takes a minimum of 3 y to establish a vine to bear fruit, meant that grapes would have been a rarer commodity than grain.

What makes the domesticated vine so desirable for largerscale production is that it is hermaphroditic, with both the male and female reproductive organs contained within a single flower, where fertilization readily occurs. The wild vine is dioecious, with separate male and female plants, so that it is dependent on the wind and, to a lesser extent, insects for pollination. Only a portion of the wild vine population—the female individuals—can produce fruit, and even then, not all flowers are pollinated. Consequently, wild vines produce far less fruit than domesticated vines. Wine making also does not make direct use of the seeds, as do beer making and bread making. Because of their bitterness, pips were usually considered waste to be discarded. In contrast, whole, unprocessed cereal grains in a bread or beer are not necessarily detrimental to the end product, and might even be considered to provide more body and taste.

Grape pressing and winemaking were generally done near where the grapes grew in antiquity, to avoid heavy transportation and conserve space within the settlement. The dense concentration of circular buildings at Shulaveri and Gadachrili would have left little room for growing grapes. Small numbers of pips might have made their way to the bottoms of the wine jars, to be disposed of later within the settlement. To date, however, no jar with seeds has been recovered from an SSC site.

Moreover, bread making and beer making require heating installations for the best results. Simply placing a mixture of ingredients under a hot sun can work, but is less reliable and efficient. Open firings around jars for beer mashing (saccharification of grain starches into sugars for fermentation) have been excavated in proto-Dynastic Egypt, ca. 3,500 BC (2, 38). Pit-firing installations associated with flat stones for possibly drying, malting, and/or baking bread or making beer are attested as early as the Pre-Pottery Neolithic period, ca. 8,700-6,500 BC, in the Near East (39). Even earlier firing installations, associated with barley starch embedded in a basalt grinding stone, have been excavated at Ohalo II, located along the southwestern shore of the Sea of Galilee and dating to the Epipaleolithic period, 23,000 y ago (40, 41). Eurasian wild grape seeds also have been reported from this site (40). Inevitably, if the processing of cereals for bread, beer, and/or another product was done nearby, some grains might have fallen into the fire or been overheated, and thus carbonized. Spent cereal grains might also have been used as fuel.

Grape fermentation does not require a heat source; in fact, a cool environment, such as a cave or burying jars underground, is best. We can conclude that bread making/beer making and winemaking occurred in different places in ancient sites, the former of which contributed to the production of masses of carbonized grains, which are well-preserved, and the latter of which resulted in low amounts of carbonized seeds.

Cereals could be dried and stored in a settlement for easy use when needed throughout the year. Grapes could be dried as raisins, but like uncarbonized pips, they generally degraded and have disappeared from the archaeological record. Grapes also can be preserved by concentrating them down into a syrup, but if this was the intended product, then pottery vessels from the SSC sites should show signs of carbon splotches due to exposure to fire on their exteriors. None do.

These considerations lead to the conclusion that the jars excavated at Shulaveri and Gadachrili, which provide chemical and archaeobotanical evidence for grape, probably originally contained wine. If their contents were high enough in alcohol, they would have provided much more than year-round sustenance for early Neolithic inhabitants. Much like Georgia's wine culture today, wine likely also served as a medicine, social lubricant, mind-altering substance, and highly valued commodity. As such, it became the focus of religious cults, pharmacopoeias, cuisines, economies, and society in general.

This "working hypothesis" (22), while buttressed by new archaeological, chemical archaeobotanical, and climatic/environmental data, is only a beginning. We may now have evidence that at least two SSC sites in Georgia, Shulaveris Gora and Gadachrili Gora, were making grape wine as much as a half millennium earlier than Hajji Firuz Tepe in Iran. However, many other regions of the Near East, especially the broad arc of mountainous terrain bordering the Fertile Crescent on its north, remain to be investigated and studied scientifically. Thus far, we have focused on jar residues from the Pottery Neolithic period, but a Pre-Pottery period preceded it, going back to ca. 10,000 BC. During the ensuing four millennia, the first permanent settlements, sustained by the founder crops, were established. Sites of this period are yet to be discovered and excavated in the SSC region of eastern Georgia, but they are well represented westward and southward in other mountainous regions.

With their extraordinary monumental architecture and artwork, Göbekli Tepe (42) and Nevali Çori (43) in the Taurus Mountains of southeastern Anatolia stand out among Pre-Pottery Neolithic sites. The domestication of three founder plants-einkorn wheat (44), chickpea, and bitter vetch-has been traced to this region. It has been proposed that wheat to make beer was the incentive that drew humans here and led to the grain's domestication. Fermentation might have been carried out in large limestone vats at Göbekli Tepe, which are the focus of ongoing chemical analyses (42). Stone bowls and goblets have also been excavated at the sites; as precursors of examples in pottery, and they were ideally suited for serving and drinking a fermented beverage. Chlorite, the stone they were made of, is a highly absorbent clay mineral that retains ancient organic compounds like pottery. The vessels are now being extracted and chemically analyzed (45).

But did the people of Göbekli Tepe and Nevali Çori limit their alcohol quaffing to wheat beer? Perhaps they experimented with wild Eurasian grape wine or honey mead. We hope to learn more about the beginnings of viniculture by the careful excavation of more archaeological sites, the fullest recovery of the micro and macro remains of our largely lost and destroyed past, and the application of the most exacting scientific techniques.

Finally, it should be noted that Jiahu in the Yellow Valley of China still has the distinction of having produced the earliest chemically confirmed grape wine in the world, as early as ca. 7,000 BC (46). This wine was probably made from a local, highsugar wild species there. However, this early Neolithic fermented beverage was not purely a grape wine, like that in the South Caucasus appears to have been, but was combined with hawthorn fruit wine, rice beer, and honey mead.

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