English · 17 September 2026
Research edition · 2.0

GHIC POSITION PAPERS

DNA-based honey authentication

Biological profiles, orthogonal evidence and reliable decisions

Executive summary

DNA analysis opens an additional biological evidence layer in honey control. It provides information on plants, bees and microorganisms that sugar-focused measurements cannot supply in the same way. GHIC advocates integrating this information into authenticity investigations, purchasing decisions and official controls, alongside proteins, chemistry and verifiable production records.

> **GHIC position:** Relevant DNA findings deserve substantive assessment and appropriate follow-up. A negative result in another method does not justify disregarding biological evidence when the methods answer different questions.

The supplied GHIC case pair illustrates the practical contribution. D339H was classified as non-authentic by DNA and contained two reported foreign enzyme targets, accompanied by atypical bee-protein findings. D340H was classified as authentic and passed the protein parameters examined. These are concrete, mutually compatible observations from different molecular layers. [7]

Honey control should use this additional information to resolve concerns more effectively. It should neither depend on a single test nor demand that a new method detect every imaginable manipulation before its useful findings are considered.

- Recognise biological profile assessment as a distinct analytical task, alongside targeted additive detection.
- Combine relevant findings through controlled sampling, transparent reporting and case-specific follow-up.
- Apply the same standards of technical quality, fair interpretation and representative references to DNA, proteomics and chemical methods.
- Build a shared reference and comparison programme that develops the evidence base while practical investigations continue.

This paper presents the scientific basis, the orthogonal case results and a practical framework for using them.

1 · What the different DNA methods establish

Approach	Useful question	Boundary
PCR, qPCR, ddPCR	Is a defined DNA target present, and in some applications how much?	Requires an appropriate marker; highly refined additives can contain too little recoverable DNA.
Metabarcoding	Which organisms are represented in selected amplified barcode regions?	Primer bias, gene-copy number and reference coverage affect relative representation.
Shotgun metagenomics	Which DNA fragments are recovered across the biological mixture?	Extraction, sequencing depth and database coverage limit detection and identification.
Authenticity classifier	Does the measured profile fit the validated authentic population?	Performance belongs to the complete workflow and model version, not to “DNA” as a technology.

Define the claim before commissioning the test

Botanical composition, geographical plausibility, a specific additive and a general authenticity classification are different measurands or decision tasks. A method validated for one cannot simply inherit authority for the others. A pollen result can help with botanical questions but does not exhaust the information on bees or microorganisms.

Paluoja and colleagues examined 369 honeys, including 266 Estonian and 103 foreign samples, and demonstrated applications to biological composition and bee pathogens. This supports the informational value of honey metagenomics. It does not independently establish the performance of the commercial MDA v2 fraud classifier. [1]

Accreditation is technically meaningful, but bounded

ISO/IEC 17025 addresses laboratory competence, impartiality and consistent operation. Evaluate the actual method and matrix in the accreditation scope, the applicable version and the statement reported. Accreditation is neither a universal test standard nor a guarantee that every possible adulteration will be detected. [13]

2 · Why honey control needs a biological evidence layer

The European Commission’s “From the Hives” action identified 147 of 320 sampled imported consignments as suspicious using the investigative approach applied. These were suspicion findings in a targeted control exercise, not a representative fraud rate for the entire honey market. They nevertheless demonstrate the need for effective follow-up and complementary control tools. [12]

Different methods expose different aspects of the product

A sugar or isotope method investigates selected chemical characteristics. DNA profiling examines biological composition. Proteomics can identify relevant enzyme targets. Production and trading records address where material originated and how lots were combined. Their value lies in answering connected questions rather than repeatedly asking one method to establish every aspect of authenticity.

The published metagenomic work by Paluoja and colleagues demonstrates access to information on honey’s biological composition and bee pathogens. The 2026 scientific comment by Pellegrin and colleagues calls for coordinated reference and comparison work for authentication applications. GHIC supports turning this into a common programme for all participating methods, with practical case evidence included from the outset. [1, 2]

A fit-for-purpose finding remains useful

Direct detection of a named syrup and assessment of a biological profile are distinct tasks. A qualitative profile can carry useful information without measuring a syrup percentage. Conversely, a highly refined, effectively DNA-free additive may leave relative DNA proportions unchanged. That scope boundary explains the need for complementary chemistry; it does not make other biological findings irrelevant.

Apply an equal evidential standard

Routine status, expensive equipment or a familiar analytical label should not determine which evidence is heard. Assess the actual question, sample, method and result. GHIC calls for evidence-led integration of biological methods and a fair examination of both concordant and discordant cases.

3 · The supplied orthogonal case pair

The controlling identifiers below reconcile the Celvia and Eurofins records. Internal shorthand labels in the underlying documents differ; identifiers and lot links are more reliable than a repeated “BG” label. The reported classifications are reproduced as analytical outcomes, not GHIC-issued certificates. [7]

Reported parameter	D339H	D340H
Eurofins sample	296-2026-00011503	296-2026-00011501
Lot	L25032026-1	L2207 / 2207
Celvia MDA v2, 10.07.2026	NON-AUTHENTIC	AUTHENTIC
Raw sequences / studied objects	10,059,764 / 273	3,627,744 / 284
α-amylase: A. oryzae	Positive	Negative
Invertase: S. cerevisiae	Positive	Negative
Other five foreign targets	Negative	Negative
MRJP1	LOW	PASSED
Bee α-amylase / α-glucosidase	ABSENT / ABSENT	PASSED / PASSED
Protein report, 23.07.2026	AR-26-TH-012014-01-THPP1	AR-26-TH-012007-01-THPP1

Proteomics: ICS SOP 520-33 (2026-02), THPP1. The issued reports specifically mark this test as accredited. They assess D339H as non-compliant and D340H as compliant for the investigated parameters. “Negative” or “absent” means not detected under the applicable assay conditions, not a claim of absolute zero.

The DNA reports describe two classification models: at least one score was below its decision threshold in D339H; both were above threshold in D340H. Actual scores were not supplied. The reports state sensitivity 89.5% and specificity 99.6%; these are laboratory-reported figures, not independently verified GHIC estimates or probabilities that an individual sample is adulterated.

> **Assessment:** One concerning and one unremarkable case show compatible DNA and protein outcomes. Two samples cannot establish general sensitivity, specificity or market prevalence. Their agreement does not identify the adulterant fraction, the entry route or fraudulent intent.

4 · What the orthogonal findings contribute

A coherent pattern across molecular layers

In D339H, the non-authentic DNA classification accompanies two identified foreign enzyme targets and atypical bee-protein findings. In D340H, both analytical layers are compatible with the respective authentic or compliant assessment. This pattern supports examining biological profiles as a useful part of honey investigations. It warrants scientific follow-up and cannot be reduced to an isolated model output. [7]

The GHIC case analysis reports relative Krona-profile shares for D339H / D340H: Apis 0.0179% / 3.3207%; bacterial groups 0.3073% / 18.4978%; Viridiplantae 57.8216% / 33.0828%. These proportions describe the reported profile denominator. They are not absolute DNA concentrations, syrup percentages or model scores; nested taxonomic levels must not be added indiscriminately. [7]

D339H produced more raw sequences than D340H. A simple “fewer reads caused the finding” explanation is therefore insufficient. Recovery, usable sequence depth and processing history remain useful comparison variables when investigating the pronounced profile differences.

Complementary does not mean identical

DNA and proteins measure different molecular layers. A purified enzyme preparation can retain detectable protein without detectable producer DNA; microbial DNA alone does not establish the corresponding enzyme preparation. The findings need not match molecule for molecule to contribute to a coherent investigation.

A focused joint follow-up

- Use controlled splits of retained samples for repeat extraction and relevant confirmation.
- Compare profiles at matched usable depth and document the reference and model versions.
- Test the interpretation against documented genuine honeys, authentic blends and known processing or admixture materials.

Interpret the combined observations jointly. Shared sample or processing effects mean that error rates should not be multiplied as if the tests were statistically independent. The pair’s evidential value is its concrete cross-method consistency.

5 · A practical DNA investigation and validation pathway

Combined finding	Required response
DNA concern + confirmed relevant protein or chemical marker	Recheck identity and analytical quality; investigate the joint hypothesis and the lot history. Escalate evidence to the competent authority where justified.
DNA concern + negative targeted panel	Keep the authenticity question open. The panel may not cover the suspected manipulation; examine extraction, processing, reference coverage and alternative chemistry.
DNA compatible + confirmed relevant foreign marker	Investigate the marker independently. A DNA model cannot clear a contradiction outside its detection capability.
Both compatible, or either inconclusive	Compatible results are limited to the methods used. An inconclusive result requires follow-up or a restricted statement, not forced clearance.

One sample history for all methods

Define the lot and sampling plan; document purchase or official sampling, seals, homogenisation, controlled splitting, storage and every handover. Allocate analysis portions and a retained counter-sample. The same product name or lot code alone does not prove that laboratories received portions of the same physical sample.

A validation programme with a defensible truth set

Use independently documented production and handling to establish authentic references across countries, bee species where in scope, seasons, floral types, honeydew, blends and processing conditions. Do not label every imported sample “fake” or every test-negative sample “authentic”. Separate training and external testing by producer and source lot to prevent information leakage.

Include blinded, traceably prepared syrup mixtures, industrial-enzyme challenges, feeding scenarios and unaltered controls. Predefine applicability, false-positive and false-negative targets, confidence intervals, inconclusive rates, repeatability, reproducibility and robustness. Report sensitivity by adulteration scenario and concentration, not as one universal number. A model update requires documented change control and relevant revalidation.

Select complementary analyses by hypothesis: appropriate isotope methods, NMR, LC-HRMS, protein markers or pollen examination. A negative C4-focused test does not exclude all C3-derived syrups. [12]

6 · Public policy, legal framework and implementation

Act within a defined evidential and legal scope

The EU Honey Directive defines honey and prohibits additions other than honey. Meeting a routine compositional value does not override that definition. Article 4 provides for implementing acts on methods by 14 June 2028; this is not a suspension of current controls. Method selection must satisfy the applicable legal hierarchy and fitness requirements. [9, 10]

Under Regulation (EU) 2017/625, Articles 34–35 govern methods and the right to a second expert opinion; Article 37 addresses official laboratories. Articles 137–138 distinguish investigation of suspected non-compliance from measures following established non-compliance. Measures must be justified for the case; neither a private model flag nor an accreditation logo automatically decides the legal outcome. [10]

Operators are responsible for compliance and traceability. Food-safety withdrawal and notification duties under Article 19 of Regulation (EC) 178/2002 must be assessed against its conditions; an authenticity issue is not automatically a demonstrated safety hazard. Deliberate fraud requires additional evidence of deception and responsibility. [11]

A concrete GHIC implementation proposal

- First 30 days: laboratories, authorities, producers and buyers agree the questions, sample plan, data fields, confidentiality rules and independence safeguards.
- Days 31–60: controlled split samples, reference materials and blinded challenges establish an auditable pilot dataset; unresolved identity or quality failures remain visible.
- Days 61–90: review concordant and discordant findings, publish a preliminary methods report and define the larger interlaboratory validation. A 90-day pilot does not equal completed validation.

Public-interest safeguards

Separate laboratory findings from commercial interests and case advocacy. Declare funding and conflicts, preserve counter-samples and correction records, and offer controlled scientific access to necessary data while protecting legitimate trade secrets. Report results proportionately and avoid unsupported public attribution of fraud.

Success means faster resolution of credible concerns, fewer unsupported accusations, better protection for honest beekeepers and more defensible purchasing and enforcement decisions. All origins and business models must face the same evidential standard.

Sources and editorial basis

Evidence cut-off: 17 September 2026. Targeted review of primary research, EU legislation and the supplied laboratory records; not a systematic review. [1, 3, 5] were assessed using indexed abstracts and bibliographic records; [2] is a scientific comment, not an experimental validation. [8] is a provider publication. Source titles remain in their original language; links are clickable.

[1] Paluoja et al. (2025). Honey bulk DNA metagenomic analysis to identify honey biological composition and monitor honey bee pathogens. npj Science of Food 9, 91. DOI ↗

[2] Pellegrin, Linsinger & Alvarellos (2026). Of bees and buzz: towards validated NGS-based methods in honey authentication. npj Science of Food 10, 129. DOI ↗

[3] Erban et al. (2021). A single honey proteome dataset for identifying adulteration by foreign amylases and mining various protein markers natural to honey. Journal of Proteomics 239, 104157. DOI ↗

[4] Takahashi et al. (2023). Investigation of Foreign Amylase Adulteration in Honey Distributed in Japan by Rapid and Improved Native PAGE Activity Staining Method. Journal of Applied Glycoscience 70, 67–73. DOI ↗

[5] Erban et al. (2026). Mining foreign proteins in honey by shotgun label-free proteomics: Implications for quality and authenticity. Journal of Proteomics, 105657. DOI ↗

[6] Wang et al. (2025). Bacillus subtilis Contributes to Amylase Production in the Honey Sac of Apis mellifera. Insects 16, 221. DOI ↗

[7] GHIC (24.07.2026). GHIC–Celvia Orthogonal Concordance Report, ET/EN. Supplied case analysis and embedded Celvia / Eurofins reports; D339H and D340H.

[8] Eurofins / Witt & Kunert. Proteomics: a new method for the detection of honey adulteration. White paper, update 21.02.2026. Source ↗

[9] European Union. Directive 2001/110/EC relating to honey, consolidated 14.06.2026; Article 4 and Annexes I–II. Source ↗

[10] European Union. Regulation (EU) 2017/625 on official controls; Articles 34–35, 37, 137–138. Source ↗

[11] European Union. Regulation (EC) 178/2002; Articles 17–19. Source ↗

[12] European Commission. EU coordinated action “From the Hives” (2021–2022): results and questions and answers. Source ↗

[13] ISO. ISO/IEC 17025:2017: General requirements for the competence of testing and calibration laboratories. Public standard overview. Source ↗

> Publisher: Global Honey Integrity Council (GHIC). Newly developed GHIC positions in five equivalent language editions. The earlier EPBA DNA paper is historical background, not a current EPBA endorsement. The case report [7] is an unpublished, two-sample GHIC analysis; its findings are paraphrased here without reproducing confidential report images or personal data. Recommendations are GHIC proposals, not pre-existing legal or laboratory requirements.