



Research article

An investigation into country of origin labeling, species authentication and short weighting of commercially sold frozen fish fillets



April M. Peterson, Gabrielle E. McBride, Seeret K. Jhita, Rosalee S. Hellberg*

Chapman University, Schmid College of Science and Technology, Food Science Program, 1 University Drive, Orange, CA, 92866, USA

ARTICLE INFO

Keywords:

Country of origin labeling
Frozen fish
Short weighting
Glazing
Species mislabeling

ABSTRACT

Proper labeling of seafood is important to prevent economic deception and protect public health. The goal of this research was to investigate prepackaged frozen fish for Country of Origin Labeling (COOL) compliance, species labeling, net weights/short weighting, and percent glaze. A total of 111 frozen prepackaged fish fillets were purchased from grocery stores in Southern California (USA). Samples were designated as COOL compliant if they displayed both procurement method and country of origin in accordance with COOL requirements. Species labeling was examined by comparing the species identified with DNA barcoding to the acceptable market names provided in the FDA *Seafood List*. Net weights and percent glaze were determined by recording the weight of each product before and after deglazing. Of the 111 samples, only 1 was noncompliant with COOL and 10 samples (9%) were short-weighted. The average percent glaze was 5%, with seven samples having >10% glaze. Most fish (95.5%) were correctly labeled with regards to species. Species substitution was discovered in two samples and three samples had unacceptable market names. The results of this study indicate high COOL compliance and minimal species mislabeling in prepackaged frozen fish fillets. However, there is a need for increased focus on short weighting and/or overglazing of frozen fish products.

1. Introduction

Americans consumed 2.4 billion kg of seafood in 2018, making the U.S. the second-largest global consumer of seafood after China (Lowther, Liddel, Yencho, & NMFS, 2020). In 2018 alone, 4.3 billion kg of seafood valued at US \$5.6 billion was commercially landed in the US, with 76.5% sold fresh/frozen for human consumption. In addition to commercial fisheries, aquaculture is an important source of seafood in the U.S. and globally. About half of the world's seafood is sourced from aquaculture, with the top three producing countries being China, India, and Vietnam. To meet the demands of consumers, the U.S. imports between 85 and 95% of seafood consumed; however, the U.S. Food and Drug Administration (FDA) only physically inspects about 2% of imported seafood, which limits their ability to identify instances of mislabeling (GAO, 2009).

Intentional mislabeling of fish species is a fraudulent act often carried out for economic gain (Silva et al., 2021). This type of fraud is challenging to detect due to the similar appearance of many fish after the morphological features have been removed during processing. Intentional mislabeling of fish and other food items is prohibited in the U.S.

according to 21 U.S.C 334: Misbranded food. In order to prevent the mislabeling of fish, the FDA recommends the use of acceptable market names given in *The Seafood List* (FDA, 2020). Despite this, previous studies conducted in the U.S. have reported the detection of species substitution as well as the use of unacceptable market names for a variety of fish species (Bosko et al., 2018; Cline, 2012; Khaksar et al., 2015; Liou et al., 2020; Mitchell and Hellberg, 2016; Pollack et al., 2018; Shokralla et al., 2015; Wang and Hsieh, 2016; Warner et al., 2013; Willette et al., 2017, 2021; Wong and Hanner, 2008). Species mislabeling not only has economic consequences but also presents health risks, including exposure to toxins such as tetrodotoxin and gempylotoxin found in pufferfish and escolar, respectively (Cohen et al., 2009; Warner et al., 2013). Fish mislabeling may also undermine the efforts of certification programs for sustainable fisheries and infringe on religious practices when non-kosher species are mislabeled as kosher species (Silva et al., 2021).

In addition to the use of acceptable market names, Country of Origin Labeling (COOL) is required for certain fresh and frozen fish fillets that are sold in the U.S. (Country of Origin labeling for Fish and Shellfish, 7 C.F.R § 60). This law requires that retailers under the Perishable Agricultural Commodities Act (PACA) provide consumers with the

* Corresponding author.

E-mail address: hellberg@chapman.edu (R.S. Hellberg).

production method and geographic origin of fresh and frozen fish fillets, steaks, and nuggets (USDA, 2020). The information must be legible to consumers and displayed in a conspicuous location. Fish that are imported into the U.S. are also subject to 19 C.F.R. § 134.11 (Country of Origin Marking Required), which requires country of origin information unless the product is exempt by law. Previous studies investigating COOL compliance among U.S. retailers have found varying levels of compliance in fresh/frozen fish, ranging from 41 to 99% (Bosko et al., 2018; Lagasse et al., 2014; Liou et al., 2020). However, there have been no studies specifically focused on COOL compliance in prepackaged frozen fish.

Additional concerns associated with frozen fish are overglazing and short weighting. A water-based glaze is commonly applied to frozen seafood products to prevent surface drying and dehydration, with adequate levels of glaze reported to be 6–10% (Vanhaecke et al., 2010). One study conducted over a five-year period in Belgium reported the average glaze on >700 samples of frozen fish marketed by a major retailer to be $8.7 \pm 2.0\%$, with a range of 2.9–16.0% (Vanhaecke et al., 2010). There are no regulations in the United States regarding the amount of glaze that can be used with seafood and excess levels of glaze are sometimes added to increase the net weight of the product artificially. This results in a short-weighted product, with customers unknowingly paying for the extra ice (NOAA, 2014). Seafood products are considered short-weighted when the difference between the advertised net weight and the actual net weight exceeds the maximum allowable variation determined by the National Institute of Standards and Technology (NIST, 2011). The FDA has received numerous complaints from other federal agencies, seafood trade associations, and the seafood industry regarding short-weighting of frozen seafood (FDA, 2009). A national survey on short weighting conducted with U.S. seafood industry members reported that half of the respondents ($n = 31$) believed that at least 71% of net weight violations in the industry were intentional (Santos et al., 2010). Ninety percent of the respondents believed that those who conduct short weighting do not feel that their actions have a negative impact further along the supply chain. Many of the survey respondents indicated frustration with regards to the lack of inspection and enforcement for short weighting. Although short weighting is a known problem in the seafood industry, there are no published studies on its prevalence in the marketplace.

The aim of this study was to investigate prepackaged frozen fish sold at the retail level for COOL compliance, species labeling, net weights/short weighting, and percent glaze. To the authors' knowledge, this is the first study to test commercially sold prepackaged frozen fish for short weighting, as well as the first combined assessment of glazing percentages, species labeling and COOL compliance in frozen fish. The results of this study are expected to reveal areas of concern with regards to labeling, overglazing and short-weighting practices for frozen prepackaged fish. This information can be used to highlight potential areas of focus for seafood inspection and enforcement efforts.

2. Materials and methods

2.1. Sample collection

Frozen fish fillets ($n = 111$) were purchased from 38 grocery stores in Southern California (USA). The stores were located within approximately 64 km of Chapman University and were in 15 different cities across Orange County, Los Angeles County, and Riverside County. All grocery stores visited for sample collection were licensed under PACA, which was verified with the PACA license search engine (https://apps.mrp.usd.gov/public_search). Only unique products were collected (i.e., no repeat sampling of the exact same product). The selection of fish was based on availability at stores and included the following 13 categories: catfish ($n = 4$), cod ($n = 15$), flounder ($n = 7$), halibut ($n = 7$), mahi-mahi ($n = 10$), orange roughy ($n = 2$), pollock ($n = 7$), salmon ($n = 15$), swai ($n = 8$), swordfish ($n = 2$), tilapia ($n = 15$), tuna ($n = 15$), and whiting ($n = 4$). A maximum of 15 fish samples was purchased per category.

2.2. COOL compliance

COOL compliance was evaluated by observing the labeling associated with each product, including tags, placards, signs, and/or packages. Photos were taken of each frozen fish package and associated signage in the store, the front and back of the packaging, the location of COOL information, the receipt, and the fillet with the packaging removed. After purchase, the fish products were transported on ice to the laboratory and held at $-20\text{ }^{\circ}\text{C}$ until deglazing and net weight determination.

2.3. Deglazing and net weight determination

The net drained weight of each sample was determined according to the AOAC official method 963.18 (a) (NFI, 2017). The fish samples were removed from the $-20\text{ }^{\circ}\text{C}$ freezer, and the net weight on the package was noted. Next, the fish was removed from the packaging, and the initial weight was collected using a MonoBlock SB32000 Weighing Balance (Mettler, Toledo) lined with aluminum foil. The contents were placed under a gentle spray of cold water using a nozzle (Peerless, PRL102, China). The fish was then agitated and sprayed with water until all the ice glaze was removed. Next, the fish was transferred to a circular No. 8 sieve (Cole-Parmer, Mentor, Ohio) inclined at an angle of $17\text{--}20^{\circ}$ for draining. Fillets weighing 0.91 kg or less were drained in a sieve with an 8 in (20.3 cm) diameter and fillets weighing more than 0.91 kg were drained in a sieve with a 12 in (30.5 cm) diameter. After draining for 2 min, the fish was immediately transferred to the scale to obtain the deglazed weight. Samples that exceeded the maximum allowed variance (MAV) according to the National Institute of Standards and Technology (NIST) standards were deemed to be short-weighted (NIST, 2011). To avoid DNA cross-contamination between samples, gloves were changed in between each sample, new aluminum foil liners were used for weighing, and tissue sampling was conducted using the interior of each fillet. The sieves were washed in between each sample using dish soap and a sponge, followed by autoclaving at $121\text{ }^{\circ}\text{C}$ for 15 min.

2.4. DNA barcoding of fish fillets

2.4.1. DNA extraction and quantification

Following deglazing, the samples were placed in the fridge at $4\text{ }^{\circ}\text{C}$ for 2–4 h to allow for partial thawing. A tissue sample (~ 10 mg) from the interior of each fillet was aseptically transferred to a 1.5 mL sterile microcentrifuge tube for use in DNA extraction. The remainder of the fillet was stored at $-20\text{ }^{\circ}\text{C}$. DNA extraction was conducted as described in Liou et al. (2020) using a DNeasy Blood and Tissue kit (Qiagen, Hilden, Germany), spin-column protocol. Lysis was performed using an Eppendorf ThermoMixer C (Hamburg, Germany) held at $56\text{ }^{\circ}\text{C}$ and 300 rpm for 3 h. DNA elution was carried out with 100 μl of preheated ($37\text{ }^{\circ}\text{C}$) AE buffer. A Biophotometer Plus (Eppendorf) was used to measure the concentration of the DNA extracted. DNA extracts with concentrations greater than 30 ng/ μl were diluted to ≤ 30 ng/ μl using AE buffer (Moore et al., 2012). The DNA extracts were held at $-20\text{ }^{\circ}\text{C}$ until use in PCR (up to 1 wk). Each set of DNA extractions included a reagent blank with no fish tissue to serve as a negative extraction control.

2.4.2. PCR amplification and confirmation

All DNA extracts were subjected to full DNA barcoding of the COI gene (655 bp), as described previously (Liou et al., 2020; Moore et al., 2012). The following components were added to each reaction tube: 8.00 μl molecular grade water, 12.5 μl 10% trehalose, one half of an OmniMix HS Lyophilized PCR Master Mix bead (Cepheid, Sunnyvale, CA), 0.25 μl of each full barcode COI primer (10 μM) and 2.00 μl of DNA template (≤ 30 ng/ μl). Thermal cycling was carried out under the following conditions: $94\text{ }^{\circ}\text{C}$ for 2 min; 35 cycles of $94\text{ }^{\circ}\text{C}$ for 30 s, $55\text{ }^{\circ}\text{C}$ for 40 s and $72\text{ }^{\circ}\text{C}$ for 1 min; and $72\text{ }^{\circ}\text{C}$ for 10 min. Samples that could not be identified using full DNA barcoding underwent mini-barcoding as described in Liou et al. (2020), with each reaction tube containing 22.0 μl molecular grade

water, one half of an OmniMix HS Lyophilized PCR Master Mix bead, 0.50 μ l of each 10 μ M COI mini-barcode SH-E primer (Shokralla et al., 2015) and 2.00 μ l of DNA template (≤ 30 ng/ μ l). Thermal cycling was carried out under the following conditions: 95 °C for 5 min; 35 cycles of 94 °C for 40 s, 46 °C for 1 min, and 72 °C for 30 s; and 72 °C for 5 min. An Eppendorf Mastercycler nexus gradient was used for all thermal cycling reactions. Amplification of PCR products was verified with pre-cast 2% agarose E-Gels (Life Technologies, Carlsbad, CA) run for 15 min on an E-Gel iBase Power System (Life Technologies), as described by Liou et al. (2020).

2.4.3. DNA sequencing

PCR purification was carried out with ExoSAP-IT (Affymetrix, Santa Clara, CA) according to the manufacturer's instructions. All samples underwent bidirectional sequencing at the GenScript facility (Piscataway, NJ) using M13 primers, BigDye Terminator v3.1 Cycle Sequencing Kit (Life Technologies) and a 3730xl Genetic Analyzer (Life Technologies). The raw data obtained from sequencing were assembled and edited with Geneious R7 (Biomatters, Ltd., Auckland, New Zealand). Consensus sequences were trimmed to the 655 bp full-length COI barcode (Handy et al., 2011) or the 226 bp SH-E mini-barcode (Shokralla et al., 2015). Full-length COI barcodes were subjected to the quality control requirements given in Handy et al. (2011): bidirectional sequences must have ≥ 500 bp and < 2 % ambiguities to pass quality control or single reads must have ≥ 500 bp and ≥ 98 % high-quality bases. The COI mini-barcode results were subjected to the quality control parameters used by Pollack et al. (2018): bidirectional sequences must have ≥ 76 % of the target length and < 2 % ambiguities to pass quality control or single reads must have ≥ 76 % of the target length and ≥ 98 % high-quality bases. The DNA barcode sequences that passed quality control were searched against the Barcode of Life Data System (BOLD) Identification Engine, Species Level Barcode Records. Sequences that could not be identified in BOLD were searched against GenBank using the Basic Local Alignment Search Tool (BLAST). The FDA Seafood List was used to determine the common name and acceptable market name for each identified species (FDA, 2020).

3. Results and discussion

3.1. COOL compliance

The 111 samples examined in this study had a high level of COOL compliance (99.1%), with only one noncompliant sample. The non-compliant sample was labeled "Hokkai cod fillet" and displayed the country of origin (China), but it did not indicate the production method. Unlike most of the other samples, this sample had a sticker-style label that may have been printed at the retail outlet and placed on the bag. The majority ($n = 107$) of the COOL compliant samples were in packages with labels that appeared to have been applied by the processors and/or they had a printed card with COOL information placed inside the packaging. A high proportion (81.0%) of the samples examined in the current study were imported, with 18 different countries of origin listed. The top seven countries declared were China ($n = 39$), USA ($n = 20$), Vietnam ($n = 17$), Taiwan ($n = 6$), Indonesia ($n = 5$), Peru ($n = 5$), and Ecuador ($n = 3$). Among the 110 samples that declared a production method, most of the fish were labeled as wild or wild-caught ($n = 80$), while the remaining samples ($n = 30$) were labeled as farmed or farm-raised.

Similar to the current study, previous research by Lagasse et al. (2014) also found a high level of COOL compliance (96.2%) for fresh and frozen fish samples sold in Baltimore city. In comparison, the Agricultural Marketing Service (AMS) reported 90% COOL compliance among retail fish and shellfish products as part of a 2016 national survey (Liou et al., 2020).

Previous studies in Southern California have reported lower rates of COOL compliance (41–77%) among fish purchased from grocery stores (Bosko et al., 2018; Liou et al., 2020). However, these studies examined

only fresh/thawed fish (Liou et al., 2020) or a combination of fresh/thawed and frozen catfish (Bosko et al., 2018). For most prepackaged frozen fish, the label is applied by the processor before it arrives at the retail outlet. In comparison, fresh/thawed fish is typically displayed at grocery store seafood counters and the retailer is responsible for proper labeling of the product. The different rates of COOL compliance indicate that there may be some confusion, lack of training, and/or lack of information provided at the retail level for the proper labeling of seafood.

3.2. Percent glaze

The average percent glaze for all 111 fish samples was $5.0\% \pm 5.5\%$, and the majority of fish samples ($n = 104$) had glaze at levels of 10% or less (Figure 1). Seven samples had $>10\%$ glaze (Table 1) and were considered overglazed based on the previously recommended maximum glazing amount of 10% (Vanhaecke et al., 2010; Seafish, 2016). Interestingly, all seven samples that were considered overglazed were labeled as wild-caught and the majority ($n = 6$) listed China as the country of origin. The highest percent glaze was found in 3 pollock/pollack samples, which had 23.0–34.5% glaze (Table 1). Direct comparisons in glaze levels were not made between fish categories due to the low sample sizes in some of the categories. Fish were sampled based on availability in the marketplace and, for some of the fish categories, only 2–4 unique products were available. Similar to the current study, Vanhaecke et al. (2010) reported that the majority of fish samples examined in their study had glaze levels of 10% or less, with 5.6% of samples having over 12% glaze. In addition to being a potentially deceptive practice, overglazing of fish can reduce the quality of the final product, for example leading to bubbling during deep frying and dilution of sauces used in cooking (Seafish, 2016). However, it is important to point out that there are no regulations regarding the percentage of glaze that can be used on frozen fish. Instead, glazing specifications may be established as part of the commercial agreement made between the buyer and the seller.

Variation in glaze levels could be due to factors such as the points in the supply chain in which glaze was applied and the type of glazing methods used (dipping vs. spraying). Dipping involves immersing the frozen product in a tank of cold water for a given time period, while spraying utilizes equipment that sprays the glazing solution over a frozen product (Soares, 2016). While dipping is relatively simple and inexpensive, it is harder to control the amount and uniformity of glaze, resulting in inconsistent glaze coverage. Regardless of the method, the amount of glaze acquired can be influenced by numerous factors, including the product size and surface area; product and glazing solution temperatures, and glazing time. Because it is difficult to obtain consistent levels of glaze, establishment of a standardized target range for % glaze on frozen seafood products may be more achievable. Additional research into glazing procedures and best practices is warranted in order to provide evidence-based recommendations for the seafood industry.

3.3. Short weighting

Short weighting was detected in 10 of the 111 fish fillets examined in this study (Table 1). Six of these samples were also overglazed (discussed above). Short weighting was detected in a variety of fish categories, including pollock, flounder, cod, tilapia, swai, and swordfish. An additional 15 samples had a deglazed weight that was less than the declared weight, but they were not considered short-weighted because they did not exceed the maximum allowable variation according to NIST (2011). Among the 10 short-weighted fish, the deglazed weight was an average of $87.1 \pm 0.9\%$ of the declared weight. In comparison, the deglazed weight for all 111 samples was an average of $101.2 \pm 5.9\%$ of the declared weight. On average, consumers were overcharged US $\$1.14 \pm 0.74$ /kg for the short-weighted samples. The most extreme case of short weighting occurred with a fish labeled as pollack (A050) whose deglazed weight was only 66.6% of the declared weight (Table 1). This sample was

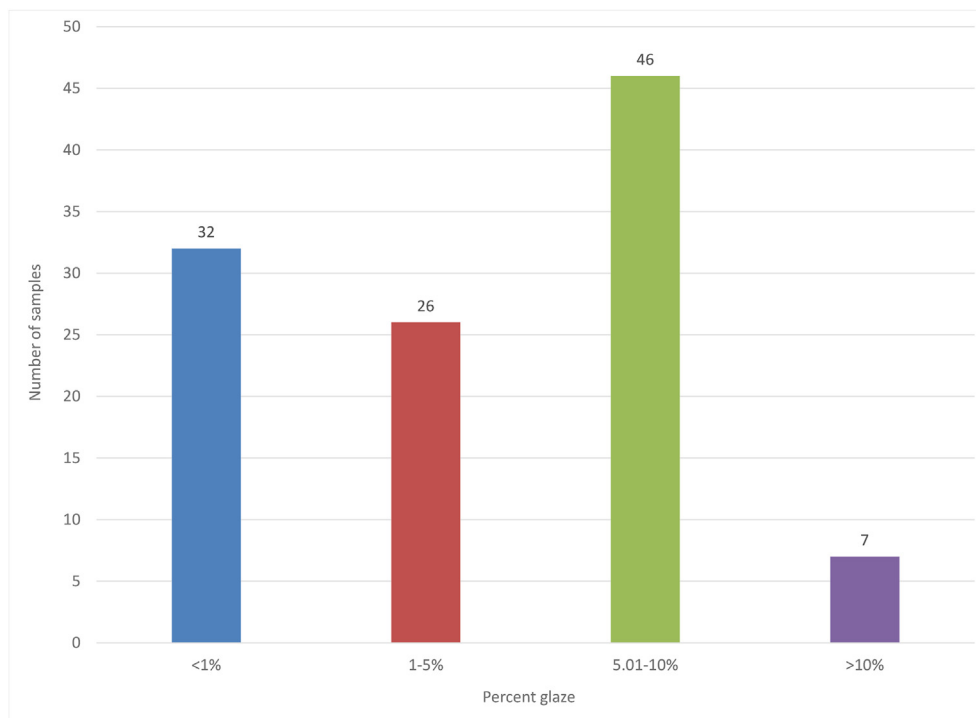


Figure 1. Percent glaze measured on prepackaged frozen fish fillets (n = 111).

Table 1. Net weight determination and % glaze for fish samples in this study determined to be overglazed (>10% glaze) and/or short-weighted. Samples that exceeded the maximum allowable variation according to NIST (2011) were considered short-weighted. Samples are listed in descending order based on percent glaze.

Sample #	Category	Product description on package	Product price (US \$/kg)	Net weight on package (g)	Glazed weight (g)	Deglazed weight (g)	Percent glaze (%)	Maximum allowable variation (g)	Detected variation ^a (g)	Deglazed weight/declared weight (%)	Price of glaze (US \$/kg) ^b
A050	pollock	pollack fillet (wild caught, China)	6.71	1016	1033	677	34.5	35.3	339	66.6	2.21
A035	pollock	pollock fillets premium (wild caught, China)	8.80	454	493	374	24.1	19.9	80	82.4	1.54
A053	pollock	pollock fillet (wild, China)	6.59	1012	1037	799	23.0	35.3	213	79.0	1.39
A038	flounder	flounder fillets premium individually vacuumed (wild caught, China)	13.21	454	487	381	21.8	19.9	73	83.9	2.13
A034	cod	cod fillets (wild caught, China)	15.41	454	474	406	14.4	19.9	48	89.4	1.63
A064 ^c	flounder	flounder fillets (wild caught, Thailand)	10.32	680	770	672	12.7	25.4	8	98.8	0.12
A081	flounder	wild Alaskan flounder (wild, China)	13.65	1600	1749	1545	11.7	49	55	96.6	0.46
A059 ^d	swai	swai fillets (farm, Vietnam)	11.00	462	467	427	8.6	19.9	39	92.4	0.84
A098 ^d	tilapia	tilapia fillets (farm-raised, Peru)	17.63	907	910	835	8.2	31.7	72	92.1	1.39
A015 ^d	flounder	flounder skinless fillets (wild caught, USA)	11.33	454	462	425	8.0	19.9	29	93.6	0.73
A001 ^d	swordfish	swordfish steaks (wild caught, Spain) on front, ahi tuna on back	17.61	412	411	391	4.9	18.1	21	94.9	0.09

^a Detected variation = net weight on package - deglazed weight.

^b Price of glaze = [100 - (deglazed weight/declared weight)] x cost of fish per kg.

^c overglazed but not short-weighted.

^d short-weighted but not overglazed

purchased for US \$6.71/kg, meaning that consumers were overcharged US \$2.21/kg.

The practice of overglazing of seafood for the purpose of artificially increasing the net weight is a fraudulent act with major potential

economic consequences. In a previous U.S. seafood industry survey on the costs of short weighting, respondents estimated that 20–40% of pollock purchased at the wholesale (import) level was less than 100% of the declared net weight, with an estimated net weight of 85–93% for

fillets (Santos et al., 2010). The estimated price paid for glaze on the short-weighted pollock imported during this time period was between US \$0.18 and 0.38/kg (Santos et al., 2010), translating to an estimated annual loss of US \$7.4–13.9 million. In comparison, pollock samples in the current study that were less than 100% of the declared weight ($n = 5$) had a wider range for the cost of glaze (US \$0.01 to \$2.21/kg). The cost of glaze for the one short-weighted tilapia sample in the current study was US \$1.39/kg, which is within the estimated range reported by Santos et al. (2010) for tilapia fillets of US \$0.25 to \$2.22/kg. The other categories of short-weighted samples from the current study had average glaze prices as follows: flounder US \$0.35 to \$0.73/kg, swordfish US \$0.09/kg, cod US \$1.63/kg, and swai US \$0.84/kg. Santos et al. (2010) did not provide short weighting or cost of glaze estimates for these categories of fish. Overall, it has been estimated that if 2% of the declared weight of seafood purchased by US consumers was ice, the annual loss to consumers would be about \$1.6 billion (Sefcik, 2011), suggesting that a small percentage of fraud could add up to billions of dollars lost. Increased inspections and enforcement surrounding short-weighted seafood products should be considered as a potential means to reduce this practice (Santos et al., 2010).

3.4. Species labeling

All 111 prepackaged frozen fish collected for this study were identified with full or mini DNA barcoding (Table 2). Most samples ($n = 106$) were identified with COI full barcoding, and the remaining five samples were identified with COI mini-barcoding. Each sample had at least one species identification in BOLD at > 98% genetic similarity, except for one sample labeled as cod that had a top mini-barcode species match to haddock (*Melanogrammus aeglefinus*) at 96% in GenBank (no sequence match in BOLD). The sequence coverage for the sample identified as haddock included the entire mini-barcode (226 bp); however, the quality was low (HQ% = 27.9%), which may explain the relatively low sequence similarity. The other four samples identified with mini-barcoding were determined to be Atlantic salmon (*Salmo salar*), walleye pollock (*Gadus chalcogrammus*), Pacific cod (*Gadus macrocephalus*)/Greenland cod (*Gadus ogac*) and yellowfin sole (*Limanda aspera*). Of the 111 samples, 67 were identified to the species level, meaning that they had a top genetic match to a single species (Table 2), and 40 samples were identified to the genus level (i.e., had a top match to more than one species from the same genus). Four tilapia samples had a top genetic match to tilapia species belonging to multiple genera in the Cichlid family (*Oreochromis* and *Coptodon*). Tilapia is difficult to identify to the species level because it is commonly cross-bred and hybridized species cannot be differentiated

with COI DNA barcoding (Dunz and Schlieven, 2013). However, the results of DNA barcoding were sufficient to confirm the labeling of these samples as “tilapia”; therefore, additional DNA testing was not conducted on these samples.

The majority of fish (95.5%) were found to be correctly labeled with regards to species and/or acceptable market name. Species substitution was revealed in two of the 111 samples, and an additional three samples had unacceptable market names (Table 3). Each of the five mislabeled samples was purchased at different stores, but two (A069 and A067) were from the same brand.

The two substituted samples consisted of (1) Kamchatka flounder (*Atheresthes evermanni*) mislabeled as halibut and (2) haddock (*Melanogrammus aeglefinus*) mislabeled as cod. Halibut labeling in the U.S. is governed by 21 CFR 102.57, which states that only two species can use the “halibut” label, Pacific halibut (*Hippoglossus stenolepis*) and Atlantic halibut (*Hippoglossus hippoglossus*). Halibut is generally a highly valued fish, however, this sample (A069) was priced at US \$8.79/kg and was the cheapest halibut sample purchased in this study. The price of the mislabeled sample was less than the average price for flounder samples purchased in this study (US \$11.57/kg). Therefore, this substitution event may have been unintentional and is possibly a result of confusion regarding proper species labeling. Although the fillets of Kamchatka flounder and halibut do not look alike (Figure 2), Kamchatka flounder and Pacific halibut are both native to the North Pacific Ocean (FishBase, 2021). Similarly, previous studies have also reported the mislabeling of flounder samples as halibut (Warner et al., 2013; Willette et al., 2017).

The mislabeling of haddock as cod may have been economically motivated due to price differences between these two types of fish (Lowther et al., 2020). However, the mislabeled haddock sample was sold at a relatively low price (US \$15.41/kg) compared to the average price of cod in this study (US \$18.96/kg). Haddock and Atlantic cod populate some of the same geographic regions (FishBase, 2021), so the haddock in this sample may have been caught in the same mass net as cod and mislabeled as cod. Along these lines, a previous market survey conducted in Ireland reported that three “cod” samples purchased from retailers were identified as haddock and one sample of “haddock” was identified as Atlantic cod (Miller and Mariani, 2010).

With the wide variety of fish species, the use of acceptable market names is essential for proper labeling of seafood in the market (FDA, 2020). As stated in the FDA Seafood List, fish should be labeled by the common name or an acceptable market name to avoid misbranding. One of the samples in this study with an unacceptable market name (A001) listed both swordfish and ahi tuna on its packaging; a sticker label with the wording “swordfish steaks” was adhered to the outside of the package

Table 2. Combined results of full and mini-DNA barcoding for fish fillets tested in this study ($n = 111$). Values are displayed as the number count.

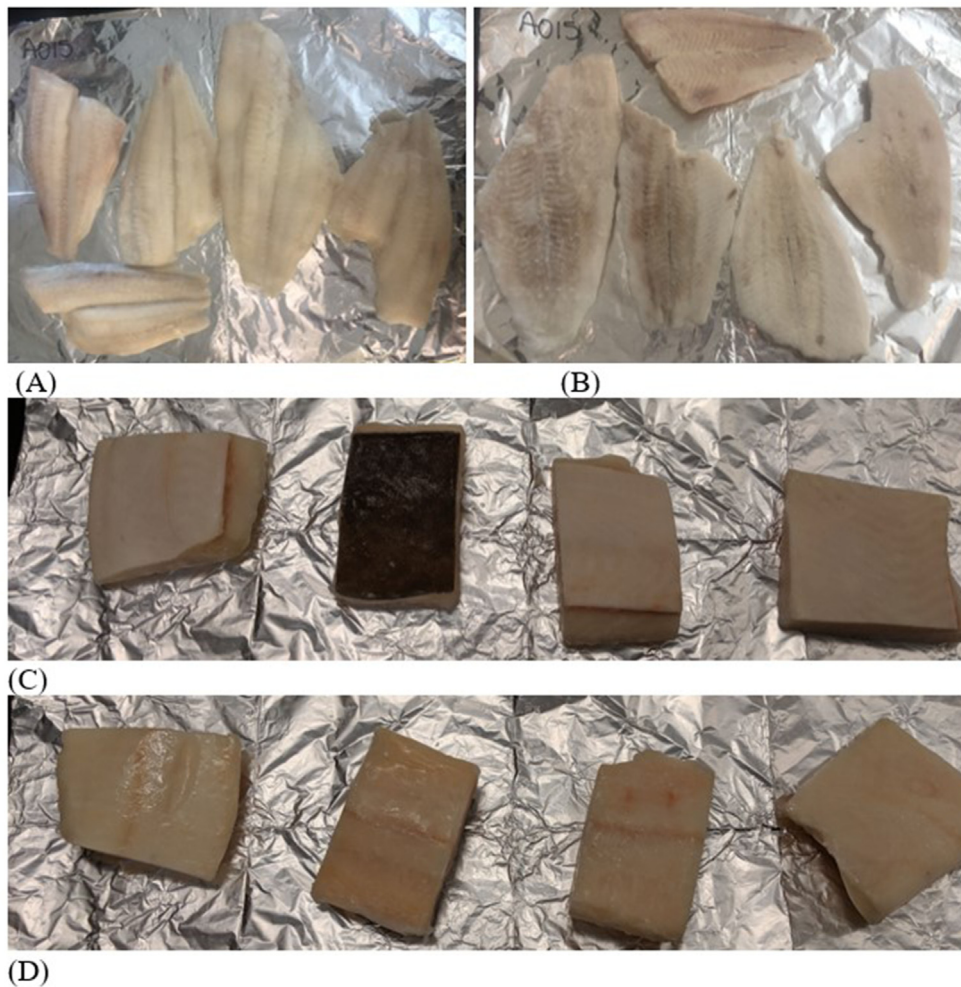
Category	Number of samples	Identified to species level	Identified to genus level	Identified to multi-genus level	Samples with species mislabeling ^a
catfish	4	3	1 (<i>Ictalurus</i>)	-	0
cod	15	5	10 (<i>Gadus</i>)	-	1
flounder	7	5	2 (<i>Limanda</i> , <i>Pleuronectes</i>) ^b	-	0
halibut	7	4	3 (<i>Hippoglossus</i>)	-	1
mahi mahi	10	10	-	-	0
orange roughy	2	2	-	-	0
pollock	7	7	-	-	2
salmon	15	15	-	-	0
swai	8	8	-	-	0
swordfish	2	2	-	-	0
tilapia	15	-	11 (<i>Oreochromis</i>)	4 (<i>Oreochromis</i> , <i>Coptodon</i>)	0
tuna	15	5	10 (<i>Thunnus</i>)	-	1
whiting	4	1	3 (<i>Merluccius</i>)	-	0
Overall	111	67	40	4	5

^a Refers to samples with species substitution or unacceptable market name.

^b One flounder sample had top genetic matches to multiple *Limanda* spp. and one sample matched multiple *Pleuronectes* spp.

Table 3. Samples in this study identified as being mislabeled due to species substitution or use of an unacceptable market name (n = 5).

Sample ID	Category	Product description on package	Expected species	Cost (US \$/kg)	Identified species: common name (scientific name)	Acceptable market name(s) other than the common name	Type of mislabeling
A034	cod	cod fillets (wild caught, China)	cod (<i>Arcotogadus borisovi</i> / <i>Arctogadus glacialis</i> / <i>Boreogadus saida</i> / <i>Eleginus gracilis</i> / <i>Gadus macrocephalus</i> / <i>Gadus morhua</i> / <i>Gadus ogac</i> / <i>Paranotothenia magellanica</i>)	\$15.41	haddock (<i>Melanogrammus aeglefinus</i>)	N/A	species substitution
A069	halibut	skinless halibut (wild, USA)	halibut (<i>Hippoglossus stenolepis</i> / <i>Hippoglossus hippoglossus</i>)	\$8.79	Kamchatka flounder (<i>Atheresthes evermanni</i>)	flounder	species substitution
A001	tuna	swordfish steaks (wild caught, Spain) on front, ahi tuna on back	tuna (<i>Thunnus</i> spp.) or swordfish (<i>Xiphias gladius</i>)	\$17.61	yellowfin tuna (<i>Thunnus albacares</i>)/blackfin tuna (<i>Thunnus atlanticus</i>)/bigeye tuna (<i>Thunnus obesus</i>)	tuna	unacceptable market name
A050	pollock	pollock fillet (wild caught, China)	N/A (no matches in Seafood List)	\$6.70	walleye pollock (<i>Gadus chalcogrammus</i>)	pollock	unacceptable market name
A067	pollock	pollock fillets (wild, China)	N/A (no matches in Seafood List)	\$4.03	walleye pollock (<i>Gadus chalcogrammus</i>)	pollock	unacceptable market name

**Figure 2.** Top and bottom sides of the fillet cuts of (A–B) Kamchatka flounder sample A069 (*Atheresthes evermanni*) mislabeled as halibut and (C–D) authenticated Pacific halibut (*Hippoglossus stenolepis*).

while a label on the inside of the package declared “ahi tuna.” This sample was identified as tuna (*Thunnus* spp.) and was deemed to have an unacceptable market name because it was labeled with conflicting species names. Two additional samples were labeled as “pollack” but identified as walleye pollock (*Gadus chalcogrammus*). According to the FDA *Seafood List*, pollack is not considered an acceptable market name for any species; however, according to FishBase (2021), “pollack” is the common name for the species *Pollachius pollachius*. The terms “pollack” and “pollock” are sometimes used interchangeably (New World Encyclopedia, 2008), which can lead to confusion in the labeling of fish species. Use of the scientific name of the species on the label would serve to reduce this confusion and promote transparency. Of note, in the current study, only about one third of the samples ($n = 35$) stated the scientific name on the package label, either as part of the ingredient list ($n = 23$) or in the product name ($n = 12$).

3.5. Combined results of mislabeling

Overall, 13 samples examined in this study had one or more labeling errors associated with COOL noncompliance, species mislabeling, and/or net weight violations. Three samples (A001, A034, and A050) had multiple labeling errors, specifically net weight violation and species mislabeling. Two of these samples A050 (misabeled pollock) and A034 (misabeled haddock) were also overglazed. These fish samples were purchased from different stores and associated with different brands. However, when considering the other samples that had labeling errors and/or overglazing, there were some common themes with regards to brand names and grocery stores. For example, samples A034 (misabeled haddock), A035 (overglazed pollock) and A038 (overglazed flounder) were from the same brand and purchased from the same store. Samples A053 (overglazed pollock) and A059 (short-weighted swai) were from the same brand and were purchased from the same chain store at two different locations; this was also the case for samples A067 (misabeled pollock) and A069 (misabeled flounder).

4. Conclusion

This is the first study to present combined data on glazing levels, net weights/short weighting, COOL compliance, and species labeling on prepackaged frozen fish fillets sold in grocery stores. The results of this study indicate a high level of compliance with COOL and accurate species labeling among prepackaged frozen fish. Relatively low levels of species substitution and unacceptable market names were noted in the samples tested in this study. Overglazing was observed in several samples, with the highest amount of glaze found in pollack/pollock samples at 34.5% glaze. Short weighting was also detected in a number of samples, most of which were also overglazed. However, the lack of a standardized target range for percent glaze on frozen seafood products makes it difficult to manage and prevent overglazing. Increased inspections and enforcement for short weighting violations may help to decrease this type of fraud. Further research into glazing, overglazing and short weighting of seafood is needed in order to increase our understanding of current practices and the extent of net weight violations.

Declarations

Author contribution statement

April M. Peterson: Conceived and designed the experiments; Performed the experiments; Analyzed and interpreted the data; Wrote the paper.

Gabrielle E. McBride, Seeret K. Jhita: Performed the experiments; Analyzed and interpreted the data.

Rosalee S. Hellberg: Conceived and designed the experiments; Performed the experiments; Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data; Wrote the paper.

Funding statement

This work was supported by the National Science Foundation, Division of Earth Sciences (NSF-EAR #1757991), and Chapman University, Schmid College of Science and Technology.

Data availability statement

Data will be made available on request.

Declaration of interests statement

The authors declare no conflict of interest.

Additional information

No additional information is available for this paper.

Acknowledgements

The authors would like to thank Carmel Williams (NOAA) for providing training on the deglazing procedure.

References

- Bosko, S.A., Foley, D.M., Hellberg, R.S., 2018. Species substitution and country of origin mislabeling of catfish products on the U.S. commercial market. *Aquaculture* 495, 715–720.
- Cline, E., 2012. Marketplace substitution of Atlantic salmon for Pacific salmon in Washington State detected by DNA barcoding. *Food Res. Int.* 45 (1), 388–393.
- Cohen, N.J., Deeds, J.R., Wong, E.S., Hanner, R.H., Yancy, H.F., White, K.D., Gerber, S.I., 2009. Public health response to puffer fish (Tetrodotoxin) poisoning from mislabeled product. *J. Food Protect.* 72 (4), 810–817.
- Dunz, A.R., Schliewen, U.K., 2013. Molecular phylogeny and revised classification of the haplotilapiine cichlid fishes formerly referred to as “Tilapia”. *Mol. Phylogenet. Evol.* 68 (1), 64–80.
- FDA, 2009. Guidance for Industry: 1991 Letter to Seafood Manufacturers Regarding the Fraudulent Practice of Including Glaze (Ice) as Part of the Weight of Frozen Seafood. Retrieved from. <https://www.fda.gov/regulatory-information/search-fda-guidance-documents/guidance-industry-1991-letter-seafood-manufacturers-regarding-fraudulent-practice-including-glaze>. (Accessed 20 January 2021).
- FDA, 2020. Guidance for Industry: the Seafood List. U.S. Food and Drug Administration. Retrieved from. <https://www.fda.gov/regulatory-information/search-fda-guidance-documents/guidance-industry-seafood-list>. (Accessed 19 January 2021).
- FishBase, 2021. A Global Information System on Fishes. Retrieved from. <https://www.fishbase.de/home.htm>. (Accessed 19 January 2021).
- GAO, 2009. Seafood Fraud: FDA Program Changes and Better Collaboration Among Key Federal Agencies Could Improve Detection and Prevention. United States Government Accountability Office. Retrieved from. <https://www.gao.gov/assets/290/287425.pdf>. (Accessed 19 January 2021).
- Handy, S.M., Deeds, J.R., Ivanova, N.V., Hebert, P.D., Hanner, R.H., Weight, L.A., Yancy, H.F., 2011. A single-laboratory validated method for the generation of DNA barcodes for the identification of fish for regulatory compliance. *J. AOAC Int.* 94 (1), 205–208.
- Khaksar, R., Carlson, T., Schaffner, D.W., Ghorashi, M., Best, D., Jandhyala, S., Amini, S., 2015. Unmasking seafood mislabeling in U.S. markets: DNA barcoding as a unique technology for food authentication and quality control. *Food Contr.* 56, 71–76.
- Lagasse, L.P., Love, D.C., Smith, K.C., 2014. Country-of-origin labeling prior to and at the point of purchase: an exploration of the information environment in Baltimore City grocery stores. *Ecol. Food Nutr.* 53 (1), 58–80.
- Liou, P., Banda, A., Isaacs, R.B., Hellberg, R.S., 2020. Labeling compliance and species authentication of fish fillets sold at grocery stores in Southern California. *Food Contr.* 112, 107137.
- Lowther, A., Liddell, M., Yencho, M., NMFS, 2020. Fisheries of the United States 2018. Current Fishery Statistics. NOAA Fisheries. Retrieved from. <https://www.fisheries.noaa.gov/resource/document/fisheries-united-states-2018-report>. (Accessed 19 January 2021).
- Miller, D.D., Mariani, S., 2010. Smoke, mirrors, and mislabeled cod: poor transparency in the European seafood industry. *Front. Ecol. Environ.* 8 (10), 517–521.
- Mitchell, J.K., Hellberg, R.S., 2016. Use of the mitochondrial control region as a potential DNA mini-barcoding target for the identification of canned tuna species. *Food Analy. Meth.* 9 (10), 2711–2720.
- Moore, M.M., Handy, S.M., Haney, C.J., Pires, G.S., Perry, L.L., Deeds, J.R., Yancy, H.F., 2012. Updates to the FDA Single Laboratory Validated Method for DNA Barcoding for the Species Identification of Fish, 4528. FDA Laboratory Information Bulletin.
- New World Encyclopedia, 2008. Pollock. Retrieved from. <https://www.newworldencyclopedia.org/p/index.php?title=Pollock&oldid=889725>. (Accessed 19 January 2021).

- NFI, 2017. Industry Guidance of Best Practices for Addressing Seafood Fraud. National Fisheries Institute. Retrieved from. <https://www.aboutseafood.com/wp-content/uploads/2018/05/BSB-Best-Practice-Guidance-November-2017.pdf>. (Accessed 19 January 2021).
- NIST, 2011. Maximum Allowable Variation for Packages. Handbook 133. Checking Net Contents of Packaged Goods. National Institute of Standards and Technology. Retrieved from. <https://www.nist.gov/document/hb133-13-finalpdf>. (Accessed 19 January 2021).
- NOAA, 2014. Illegal, Unregulated, and Unreported Fishing and Seafood Fraud. Retrieved from. <https://www.iuufishing.noaa.gov/>. (Accessed 19 January 2021).
- Pollack, S.J., Kawalek, M.D., Williams-Hill, D.M., Hellberg, R.S., 2018. Evaluation of DNA barcoding methodologies for the identification of fish species in cooked products. *Food Contr.* 84, 297–304.
- Santos, J., Roheim, C., Durham, K., 2010. Final Report: Improving the Economic Integrity of the U.S. Seafood Industry: Analysis of the Costs of Short Weighting. National Fisheries Institute. University of Rhode Island.
- Seafish, 2016. Glaze (FS94_07_16). Retrieved from. https://www.seafish.org/media/publications/FS94_07_16_Glazing.pdf. (Accessed 17 June 2020).
- Sefcik, D., 2011. Seafood fraud - where are we now? *Natl. Inst. Stand. Technol. Newslett.: Weights Meas. Connect.* 2 (7), 1–2. Retrieved from. <https://www.nist.gov/publications/seafood-fraud-where-are-we-now>. (Accessed 19 January 2021).
- Shokralla, S., Hellberg, R.S., Handy, S.M., King, I., Hajibabaei, M., 2015. A DNA mini-barcoding system for authentication of processed fish products. *Sci. Rep.* 5, 15894.
- Silva, A.J., Hellberg, R.S., Hanner, R.H., 2021. Seafood fraud. In: Hellberg, R.S., Everstine, K., Sklare, S.A. (Eds.), *Food Fraud: A Global Threat with Public Health and Economic Consequences*. Academic Press/Elsevier, San Diego, pp. 109–137.
- Soares, N.F., 2016. Updating Use of Glazing in the Frozen Seafood Industry. *Food Quality & Safety*. Retrieved from. <https://www.foodqualityandsafety.com/article/updating-use-of-glazing-in-the-frozen-seafood-industry/>. (Accessed 19 January 2021).
- USDA, 2020. Country of Origin Labeling (COOL). U.S. Department of Agriculture Agricultural Marketing Service. Retrieved from. <https://www.ams.usda.gov/rules-regulations/cool>. (Accessed 19 January 2021).
- Vanhaecke, L., Verbeke, W., De Brabander, H.F., 2010. Glazing of frozen fish: analytical and economic challenges. *Anal. Chim. Acta* 672 (1-2), 40–44.
- Wang, D., Hsieh, Y.-H.P., 2016. The use of imported pangasius fish in local restaurants. *Food Contr.* 65, 136–142.
- Warner, K., Timme, W., Lowell, B., Hirshfield, M., 2013. Oceana Study Reveals Seafood Fraud Nationwide. Retrieved from. <https://oceana.org/reports/oceana-study-reveals-seafood-fraud-nationwide>. (Accessed 19 January 2021).
- Willette, D.A., Simmonds, S.E., Cheng, S.H., Esteves, S., Kane, T.L., Nuetzel, H., Barber, P.H., 2017. Using DNA barcoding to track seafood mislabeling in Los Angeles restaurants. *Conserv. Biol.* 31 (5), 1076–1085.
- Willette, D.A., Esteves, S.C., Fitzpatrick, B., Smith, M.L., Wilson, K., Yuan, X., 2021. The last mile challenge: certified seafood and federal labeling laws out of sync at the end of the supply chain in Los Angeles, California. *Mar. Pol.* 125, 104380.
- Wong, E.H.K., Hanner, R.H., 2008. DNA barcoding detects market substitution in North American seafood. *Food Res. Int.* 41 (8), 828–837.