Food Control 94 (2018) 38–47

Contents lists available at ScienceDirect

Food Control

journal homepage: www.elsevier.com/locate/foodcont

Study of fish products in Metro Vancouver using DNA barcoding methods reveals fraudulent labeling

Yaxi Hu[a], Shr Yun Huang[a], Robert Hanner[b,c], Julia Levin[d], Xiaonan Lu[a,e,*]

[a] Food, Nutrition and Health Program, Faculty of Land and Food Systems, The University of British Columbia, Vancouver, V6T 1Z4, BC, Canada
[b] Department of Integrative Biology, University of Guelph, Guelph, N1G 2W1, ON, Canada
[c] Biodiversity Institute of Ontario, University of Guelph, Guelph, N1G 2W1, ON, Canada
[d] Oceana Canada, Toronto, M5C 1C4, ON, Canada
[e] Peter Wall Institute for Advanced Studies, The University of British Columbia, Vancouver, V6T 1Z2, BC, Canada

ARTICLE INFO

Keywords:
DNA barcoding
DNA mini-barcoding
Species substitution
Food fraud
Regulation

ABSTRACT

The complexity of seafood supply chains and fish naming systems has rendered the traceability of a seafood product challenging if not impossible, creating loopholes for intentional or unintentional illicit practices that erode the market transparency and integrity. DNA barcoding has been extensively applied to evaluate the integrity of seafood markets worldwide. However, little information is available to comprehensively assess the fish market integrity in Metro Vancouver, the 3rd largest metropolitan city in Canada. Therefore, we conducted a market survey by collecting 285 finfish samples from grocery stores, sushi bars and non-sushi restaurants in Metro Vancouver and used DNA barcoding and DNA mini-barcoding to analyze the compliance of product market names with allowable species in accordance with the Canadian Fish List. On average 25% of the samples were mislabeled. Restaurants had the highest mislabeling rate (*i.e.* 29%), followed by grocery stores (*i.e.* 24%), while sushi venues (*i.e.* 23%) had the least mislabeling incidence. Among the mislabeled products, snappers presented the highest mislabeling rate (*i.e.* 91%). Evidence of the various motivations of the mislabeling, including intentional substitution using less expensive species, purposed mislabeling of by-catch or illegal, unreported and unregulated fishing products, and unintentional misidentification or misuse of dialects and vernaculars were all observed. To protect the integrity of the seafood supply chain, several recommendations on the labeling standards are discussed: harmonize the accepted common names with major trading countries; enforce labels to include scientific names for fish products; disapprove the use of vague multispecies generic names; and require labels to provide information of geographical origins and catching/farming methods. These actions have contributed to the significant improvement of seafood market integrity in the European Union and are anticipated to be effective in Canada.

1. Introduction

Fish is one of the major protein foods with an annual consumption of over 100 million tonnes worldwide (Food and Agriculture Organization, 2016). Canada had an annual per capita fish consumption of 22.5 kg during 2013–2015 and ranks 21st in fish harvesting by weight globally (Food and Agriculture Organization, 2016). Canada is a leading fishing nation, with the leading fish species harvested/produced in Canada including salmon, herring, hake, redfish and haddock (Fisheries and Oceans Canada, 2012). Although the fish species harvested or produced in Canada are limited by geographical factors, more than 900 species of fish are available in the market (Canadian Food Inspection Agency, 2017a).

Canada is among the top ten fish exporters in the world with 85% of its harvest being exported (Food and Agriculture Organization, 2016), but it is also heavily dependent upon seafood imports which constitute approximately 80% of the seafood available in Canadian markets (Oceana Canada, 2017). Interestingly, fish imported into Canada have often been harvested or farmed domestically in Canada but processed elsewhere, commonly in China. In a typical fish supply chain, four or even more independent countries could be responsible for the different steps in fish processing, including harvesting/farming, primary processing (*e.g.* freezing, filleting and gutting), secondary processing (*e.g.* canning and breading), and packaging. Finally, the packaged fish products could be sold domestically or exported to other countries and could include the country where the fish was harvested in the first

* Corresponding author. Food, Nutrition and Health Program, Faculty of Land and Food Systems, The University of British Columbia, Vancouver, V6T 1Z4, BC, Canada.
E-mail address: xiaonan.lu@ubc.ca (X. Lu).

https://doi.org/10.1016/j.foodcont.2018.06.023
Received 9 March 2018; Received in revised form 12 June 2018; Accepted 13 June 2018
Available online 18 June 2018
0956-7135/ © 2018 Elsevier Ltd. All rights reserved.

place. Within such a complex and fragmented supply chain, effective and efficient tracing and tracking of products is problematic, and implementation difficult (Wang, Van Fleet, & Mishra, 2017). One of the consequences associated with such complicated seafood supply chains is the high mislabeling rate (intentional and unintentional practices) of fish products around the world, including the United States (US) (Warner, Timme, Lowell, & Hirshfield, 2013), China (Xiong, D'Amico et al., 2016), Italy (Armani et al., 2015), the United Kingdom (Vandamme et al., 2016), and South Africa (Cawthorn, Steinman, & Witthuhn, 2012). Intentional substitutions of high quality fish with less expensive fish, or selling fish from illegal, unreported and unregulated fishing (IUU) operations are common. Additionally, unintentional mislabeling due to the misidentification of fish species or ambiguous labeling regulations were also recognized. Regardless of the motivations for mislabeling, lack of integrity in global fish markets lead to: 1) economic damage to consumers and responsible fish industries; 2) diminished consumer trust in governmental agencies and the food industry; 3) potential health threats to susceptible populations from mislabeling (*e.g.* histamine forming scombroid fish substituted for non-scombroid fish); 4) the creation of a legal market for products of IUU fishing; and 5) impeding consumers from making informed decisions to support sustainable fisheries (Naaum, Warner, Mariani, Hanner, & Carolin, 2016).

The high prevalence of fish mislabeling can be attributed in part to the complexity of the supply chain, but inability to monitor and detect species substitution has provided opportunities for unscrupulous marketers (Reilly, 2018). Other factors, such as the inability to visually identify fish like rockfish to the species level makes identification difficult, even for fisherman. Furthermore, the removal of species-specific features such as the head, fins, skin and scales, during fish processing greatly complicates reliable visual identification. Lastly, the differences in vernacular and common local names of fishes, having the same common name for different fishes (*e.g.* redfish, snapper, salmon, trout, and pollock) in the domestic and international markets, and lack of knowledge or appropriate use of the proper scientific and internationally recognized names for fishes have created discrepancies in the standardization of labeling in the international fish trade (Cawthorn & Mariani, 2017). Fishes are unique and have inherent properties favored by unscrupulous industries. Therefore, to protect consumers and the integrity of the fish industry, steps should be taken to reduce seafood mislabeling and the resulting consumer fraud.

Over the past decade, several significant amendments in government standards have been implemented. For example, European Union (EU) requires all the fish products to be labeled with sufficient details to enable traceability (*i.e.* scientific name, common market name, wild/farmed, harvest/production areas and harvesting/production methods) (Council of the European Union & European Parliament, 2013). In the US, the Food and Drug Administration (FDA) has adopted a DNA barcoding method to replace their previous fish identification analytical method based on isoelectric focusing (Handy et al., 2011). These changes have been made to improve the integrity of fish marketing. In addition, massive media coverage on fish fraud and mislabeling issues has aroused the attention of consumers and put pressure on fish processors, distributors, restaurants and retailers to accurately label the fish being sold. According to Mariani et al. (2014), media pressure played an important role in reducing cod mislabeling rate from 35% to 0% in grocery stores in Dublin, Ireland between 2009 and 2012, indicating that actions taken by EU were effective. A transnational study conducted in Western Europe from 2013 to 2014 unveiled a significant improvement in the EU fish market with less than a 5% mislabeling rate (Mariani et al., 2015), although the situation in some Southern European countries was not as promising as this due to some cultural variations (Tinacci et al., 2018). However, less encouraging results were observed in an Oceana nationwide study conducted in the US from 2010 to 2012 with an average mislabeling rate of 33% (Warner et al., 2013). In more recent studies that analyzed commonly consumed sushi species in Los Angeles (Willette et al., 2017), salmon products in grocery stores and restaurants (Warner et al., 2015), and sushi and wholesale/retails samples in three US metropolitan areas (Khaksar et al., 2015), mislabeling rates were 47%, 43% and 16.3%, respectively. Compared with the lower fish mislabeling rate in the EU countries, mislabeling in US markets is attributed to the less strict enforcement of labeling regulations (Mariani et al., 2015).

The labeling regulations for fish products in Canada require no detailed information regarding the identity of the fish themselves (*e.g.* geographic origins, wild or farmed, and catching or farming methods), while the US requires the addition of labeling the fish as wild caught or farmed. Labeling the seafood product with a common name listed by US FDA or Canadian Food Inspection Agency (CFIA) and the country of origin (*i.e.* country where the fish was last transformed) is acceptable in the corresponding country. However, inconsistencies between the use of common name of fishes accepted by the US and Canada are not unusual, even though the US is the largest importer and exporter of Canada's seafood (Agriculture and Agri-Food Canada, 2016; Oceana Canada, 2017). Such discrepancy has resulted in confusion in regional markets since Pacific rockfish (*Sebastes* sp.) can be legally sold as Pacific red snapper in Washington, Oregon and California – though not in Canada – causing confusion with the more expensive Gulf of Mexico red snapper (*Lutjanus* sp.) (Jacquet & Pauly, 2008). Although the latest Food Safety Modernization Act (FSMA) issued by the US FDA has included a chapter for protecting food against intentional adulteration (US Food and Drug Administration, 2013), food fraud (*i.e.* economically motivated adulteration) is not covered by the act (US Food and Drug Administration, 2017a) unless the species substitution would result in adverse health impacts, such as substitution of a scombroid (histamine forming species) for one that does not form histamine, or in cases where species substitution would result in allergenic reactions. The US Food Drug and Cosmetic Act (FDCA), including provisions in FSMA, require that fish be labeled as an allergen and species name be included on product labels under the amendment to the FDCA in the Food Allergy Labeling and Consumer Protection Act (FALCPA) in 2004. Section 403 of the FDCA also prohibits misbranding of food, and this includes species substitution. However, based only on a generic common name and country of origin provided on the label, mislabeling of seafood are difficult to control. Similarly in Canada, fish industries are monitored according to the “Fish Inspection Program Sampling Procedures”. This standard explicitly states that the fish will be subjected to sensory, chemical indicator, package integrity and net content analyses, container integrity analysis, and/or microbiology and chemistry analyses (Canadian Food Inspection Agency, 2013). The focus of the fish inspection program is to eliminate physical, chemical and biological safety concerns instead of monitoring the authenticity or legality of the fish industry. Although the “Consumer Packaging and Labelling Act” is intended to provide consumers with truthful and not misleading labels, with limited information provided on the label and insufficient inspection, effective control of fish mislabeling is challenging.

In the US, dozens of articles have been published investigating the integrity of seafood markets, but only three research articles could be identified to date analyzing the mislabeling rates of Canadian seafood markets in peer-reviewed journals (Hanner, Becker, Ivanova, & Steinke, 2011; Naaum Amanda & Hanner, 2015; Wong & Hanner, 2008), in addition to one formal report generated by Oceana Canada (Levin, 2017). Moreover, the majority of the samples (n = 719) collected in those studies was from the Eastern Canada in Ontario and Montreal or the US, while only 64 samples were collected from the Western Coast of Canada in Vancouver. In addition, most of the market survey studies conducted in the US and Canada used DNA barcoding method, identifying different fish based upon the sequence of a 650-bp mitochondrial COI DNA fragment, also believed to be the DNA fingerprint of a biological species (Ward, Hanner, & Hebert, 2009). For some processed food products, much of the DNA has been degraded into sequences of less than 300–400 bp; therefore studies for definitive identification of

fish usually exclude highly processed food samples (*e.g.* canned or dried fish) due to a failed DNA amplification step (Hanner et al., 2011). To circumvent this drawback of DNA barcoding, an alternative barcoding method, recognized as mini-barcoding, has been developed (Hajibabaei et al., 2006). By designing universal primers focusing on a shorter DNA sequence, fish identity in highly processed food products could also be determined (Shokralla, Hellberg, Handy, King, & Hajibabaei, 2015).

Therefore, to have a better understanding of the integrity of the seafood market on the west coast of Canada and given that the last survey conducted in Vancouver was nearly a decade ago, we conducted a more comprehensive survey study with a larger sample size focusing on Metro Vancouver. Less than three-hundred fish samples were collected from grocery stores, sushi bars and non-sushi restaurants located in multiple Metro Vancouver cities. To the best of our knowledge, this was the first seafood market survey study focused specifically on Metro Vancouver by applying both DNA barcoding and DNA mini-barcoding methods.

2. Material and methods

2.1. Collection of fish samples

Fish species that are of high regional significance or had high mislabeling rates in North America (Cline, 2012; Hanner et al., 2011; Lowenstein et al., 2010; Wong & Hanner, 2008) were selected as target species for the current study. Grocery stores, sushi bars and non-sushi restaurants in Metro Vancouver, specifically Vancouver, Burnaby, Coquitlam, Surrey, Langley and North Vancouver, were selected randomly or based on the availability of the fish of interest. A total of 285 samples were acquired between September 2017 and February 2018. Samples purchased from sushi bars or non-sushi restaurants were ordered for take-out and the identity of fish species was doubly confirmed with staff in the restaurants. Samples were transferred to the lab immediately and stored at 4 °C and sub-sampled within 24 h. Sub-sampling was conducted by cutting a small piece of muscle tissue (*c.a.* 1 mm cube) from the center of the sample and storing this in 95% ethanol at −20 °C until DNA extraction. Cross-contamination was prevented by treating tools with ELIMINase (Decon Laboratories, USA) between samples to eliminate DNA and DNase residues carried over from the previous sample. Tissue from each sample of at least 1 cm^3 tissue was covered with plastic wrap for each sample and stored at −20 °C as a backup.

2.2. Total DNA extraction

Total DNA extraction was performed using Qiagen DNeasy Blood & Tissue kits (Qiagen Inc., Mississauga, ON, Canada) following the instructions of the manufacturer with minor modifications. Briefly, fish muscle tissue stored in 95% ethanol were mixed with 180 μL of ATL lysis buffer and 20 μL of proteinase K and incubated at 56 °C overnight with constant shaking at 220 rpm. DNA extraction was then performed according to the instruction manual and was recovered with 100 μL of elution buffer.

2.3. Amplification of the DNA barcode regions

FISH-BOL recommended M13-tailed fish-cocktail primers targeting at a 652-bp region on the mitochondrial COI gene (Hanner et al., 2011) are listed in Table 1. The PCR reaction mixtures contained 1.25 μL of 10× Platinum Taq PCR Buffer, 0.625 μL of 50 mM $MgSO_4$, 0.0625 μL of 10 mM dNTPs, 0.125 μL of each forward and reverse primers working solutions, 0.05 μL of Platinum Taq polymerase, 3 μL of fish DNA, and 7.26 μL of sterile water to achieve a final volume of 12.5 μL. Forward primer working solution contains 5 μM of VF2_t1 and 5 μM of Fish F2_t1, while reverse primer working solution contains 5 μM of FR1d_t1 and 5 μM Fish R2_t1. The PCR thermal cycling program consisted of an initial denaturation at 95 °C for 4 min, 5 cycles of denaturation at 95 °C for 30 s, annealing at 50 °C for 30 s, and elongation at 72 °C for 1 min, and 35 cycles of denaturation at 95 °C for 30 s, annealing at 54 °C for 30 s, and elongation at 72 °C for 1 min. PCR was concluded by a final elongation at 72 °C for 5 min and held at 4 °C before being examined using 1% electrophoresis agarose gel stained with Red Safe DNA dye.

Samples that cannot be amplified using fish-cocktail primers were subjected to amplification using mammal-cocktail primers and mini-barcode primers (Table 1).

2.4. DNA sequencing analysis

Amplicons from successful PCR were purified (QIAquick PCR purification kit, Qiagen Inc., Mississauga, ON, Canada) following the manufacturer's instructions and then sent to UBC Sequencing and Bioinformatics Consortium (NAPS) for Sanger sequencing. The trace files acquired from Sanger sequencing were analyzed using CodonCode Aligner 7.1.2 (CodonCode Corporation, Dedham, MA) to obtain DNA sequences. DNA sequences with a length of > 550 bp for DNA barcode and > 100 bp for mini-barcode were searched on Barcode of Life Database (BOLD, http://www.boldsystems.org/index.php/IDS_IdentificationRequest). The species identified with a sequence similarity of > 98% was considered as a match for a particular fish sample. Fish were identified by comparing the name labeled on the product in the market with the acceptable common names on the CFIA Fish List (Canadian Food Inspection Agency, 2017a) corresponding to the scientific name retrieved after the analysis on BOLD. For fish species absent in the CFIA Fish list, the FDA Seafood List (US Food and Drug Administration, 2017b) was referred to. Samples were considered to be mislabeled if the market names did not match the accepted common names for a specific fish. The motivations of the mislabeling practices were analyzed case-by-case.

2.5. Statistical analysis

To analyze whether sampling source, product form, appearance, flesh color or if the fish had been raw or cooked could impact the mislabeling practice, a generalized linear regression model with binary distribution was constructed using MATLAB 2017b (Mathworks, Natick, Massachusetts, USA). Properties were recognized as being significant if a $p < 0.05$ was acquired.

3. Results and discussion

3.1. Overall results

The number and composition of samples collected in Metro Vancouver area are summarized in Fig. 1 and Table S1. Among the 285 samples tested, 272 samples were successfully sequenced using Sanger sequencing technique for their DNA barcode, while 9 samples were analyzed for their DNA mini-barcode. There were four samples that failed both DNA barcoding and mini-barcoding methods. All the detailed information about the samples (specimen) and the sequences has been submitted to the BOLD system under a project entitled FISHV. Although various universal primers for DNA mini-barcodes have been designed to analyze highly processed foods, relatively unsatisfactory success rate with DNA amplification (~70%) has commonly been reported (Chin Chin, Adibah, Danial Hariz, & Siti Azizah, 2016; Mitchell & Hellberg, 2016). Three possible reasons for the failure of mini-barcoding for those samples are 1) DNA in the sample has been denatured into even smaller fragments leaving an insufficient amount of DNA template, 2) PCR inhibitors (*e.g.* lipids) presented in the samples hindered the DNA amplification, and 3) Sanger sequencing technique is less suitable for analyzing DNA sequence shorter than 250 bp (McGill University, 2010). To analyze highly denatured DNA in processed foods, quantitative PCR using species specific primers or probes targeting < 100-bp DNA region might be more appropriate (López-

Table 1
Primers for fish barcode and mini-barcode DNA regions.

Name	Primer sequence 5'→3'	Reference
	M13-tailed Fish-Cocktail DNA-barcode primers	25
VF2_t1	TGTAAAACGACGGCCAGTCAACCAACCACAAAGACATTGGCAC	
FishF2_t1	TGTAAAACGACGGCCAGTCGACTAATCATAAAGATATCGGCAC	
FishR2_t1	CAGGAAACAGCTATGACACTTCAGGGTGACCGAAGAATCAGAA	
FR1d_t1	CAGGAAACAGCTATGACACCTCAGGGTGTCCGAARAAYCARAA	
	M13-tailed Mammal-Cocktail DNA-barcode primers	25
LepF1_t1	TGTAAAACGACGGCCAGTATTCAACCAATCATAAAGATATTGG	
VF1_t1	TGTAAAACGACGGCCAGTTCTCAACCAACCACAAAGACATTGG	
VF1d_t1	TGTAAAACGACGGCCAGTTCTCAACCAACCACAARGAYATYGG	
VF1i_t1	TGTAAAACGACGGCCAGTTCTCAACCAACCAIAAIGAIATIGG	
LepR1_t1	CAGGAAACAGCTATGACTAAACTTCTGGATGTCCAAAAAATCA	
VR1d_t1	CAGGAAACAGCTATGACTAGACTTCTGGGTGGCCRAARAAYCA	
VR1_t1	CAGGAAACAGCTATGACTAGACTTCTGGGTGGCCAAAGAATCA	
VR1i_t1	CAGGAAACAGCTATGACTAGACTTCTGGGTGICCIAAIAAICA	
	Mini-M13 tailed DNA mini-barcode primers	28
UniMiniBar_F	TGTAAAACGACGGCCAGTTCCACTAATCACAARGATATTGGTAC	
UniMiniVar_R	CAGGAAACAGCTATGACGAAAATCATAATGAAGGCATGAGC	
	M13 primers for sequencing the DNA-barcode amplicons	25
M13F	TGTAAAACGACGGCCAGT	
M13R	CAGGAAACAGCTATGAC	

Andreo, Aldeguer, Guillén, Gabaldón, & Puyet, 2012). Alternatively, next-generation sequencing techniques have the potential to be applied in routine food fraud analysis with the improvement in the hardware performance (Giusti, Armani, & Sotelo, 2017).

DNA Sanger sequencing results with > 550 bp nucleotides for DNA barcode region and > 100 bp for DNA mini-barcode region were compared with the DNA database on BOLD. Although a definitive species level identification was not achieved for some of the samples, determination of the authenticity or mislabeling of samples was impacted slightly. The labeling accuracy of each fish sample was determined according to the CFIA fish list. All fish on the market should be labeled with a CFIA accepted common name instead of using vernaculars for the corresponding biological species determined by their DNA barcodes, and biological species that are not included in the CFIA fish list should not enter Canadian fish markets (Canadian Food Inspection Agency, 2017b). Any fish for which the DNA sequence did not match the CFIA list were considered to be mislabeled. A total of 25% of the samples tested were identified as mislabeled and the detailed information for theses samples is summarized in Fig. 2 and Table 2. This indicates that there has been no significant improvement in the integrity of fish labeling in the Vancouver retail and restaurant markets over the past decade and since the last study (Hanner et al., 2011). For the mislabeled samples, 89% of them had unacceptable common names for a CFIA listed fish species; while 8 samples were identified as biological species that are not allowed for sale in Canada unless a further update of the CFIA fish list is released.

To investigate if other properties could impact mislabeling, samples were categorized based on five parameters, namely raw/cooked, source of samples (*i.e.* sushi bar, grocery store and non-sushi restaurant), appearance (*e.g.* plain fillet, seasoned, and covered with breading or other ingredients), product form (*e.g.* whole fish, fillet, chunk, and chopped), and color of the flesh [e.g. red and light color (pink/white)]. Generalized linear regression model with binomial distribution, a logistic analysis, was constructed to analyze the significance of these five factors on mislabeling practices and the results are summarized in Table 3. Providing a significance level of $\alpha = 0.05$, the product form and color of the flesh had a statistically significant impact on the mislabeling issues, while no significant difference in mislabeling rate was observed for samples collected from different sources. Products composed of fish muscle of a lighter color flesh and chopped muscle tissue were more prone to illicit practices compared to red or fillet samples, and this aligns with our common sense that fish with light color flesh is less distinctive than fish with colorful flesh, such as sockeye salmon, although we did observe mislabeling of salmon products in this study. Fish products with extensive removal of morphological attributes were more prone to species substitution. However, whole fish also had a higher mislabeling rate compared with fish fillets, which could be attributed to a bias in our sampling method – a much smaller number of whole fish samples with targeting species were collected compared to the other forms. In addition, the impact of raw/cooked and the appearance attributes of fish products was not statistically significant. Again, this could be explained by the bias in our sampling method. Raw fish samples and breaded samples were collected mainly from sushi bars and grocery stores with a focus on particular species that have been historically associated with mislabeling/fraud incidences, and this would make the significance of these two factors in the analysis less reliable. Nevertheless, the level of species substitution and motivations for mislabeling was different depending upon the sources from which the samples were purchased.

3.2. Sushi bars

A total of 107 sushi samples purchased from 39 sushi bars were successfully sequenced with a successful rate of 99%. The mislabeling rate of 22% was the lowest compared with that of non-sushi restaurants and grocery stores (Fig. 2). The mislabeling rate in sushi bars was much lower in Metro Vancouver compared to previous studies conducted in Eastern Canada, US and Europe (Hanner et al., 2011; Oceana, 2015; Warner et al., 2013). Located on the coast of northern Pacific Ocean, Metro Vancouver has an easy access to abundant resources of high quality sushi fish, and this could contribute to the high degree of fish authenticity in sushi bars in this community. However, the level of integrity of sushi market in Vancouver is still not satisfactory.

Although substitution of Pacific wild salmon by Atlantic farmed salmon was not observed in Metro Vancouver, fraudulent snapper and red snapper marketing practices were prevalent (*i.e.* 100%), which conforms with the high mislabeling rate in the previous studies (Hanner et al., 2011; Warner et al., 2013). All but one of the snapper and red snapper sushi tested in this study were identified as tilapia, a much cheaper fish species compared with snappers, indicating intentional substitution for economic gain. One of the red snapper samples was identified as silver seabream. The intention behind this mislabeling case

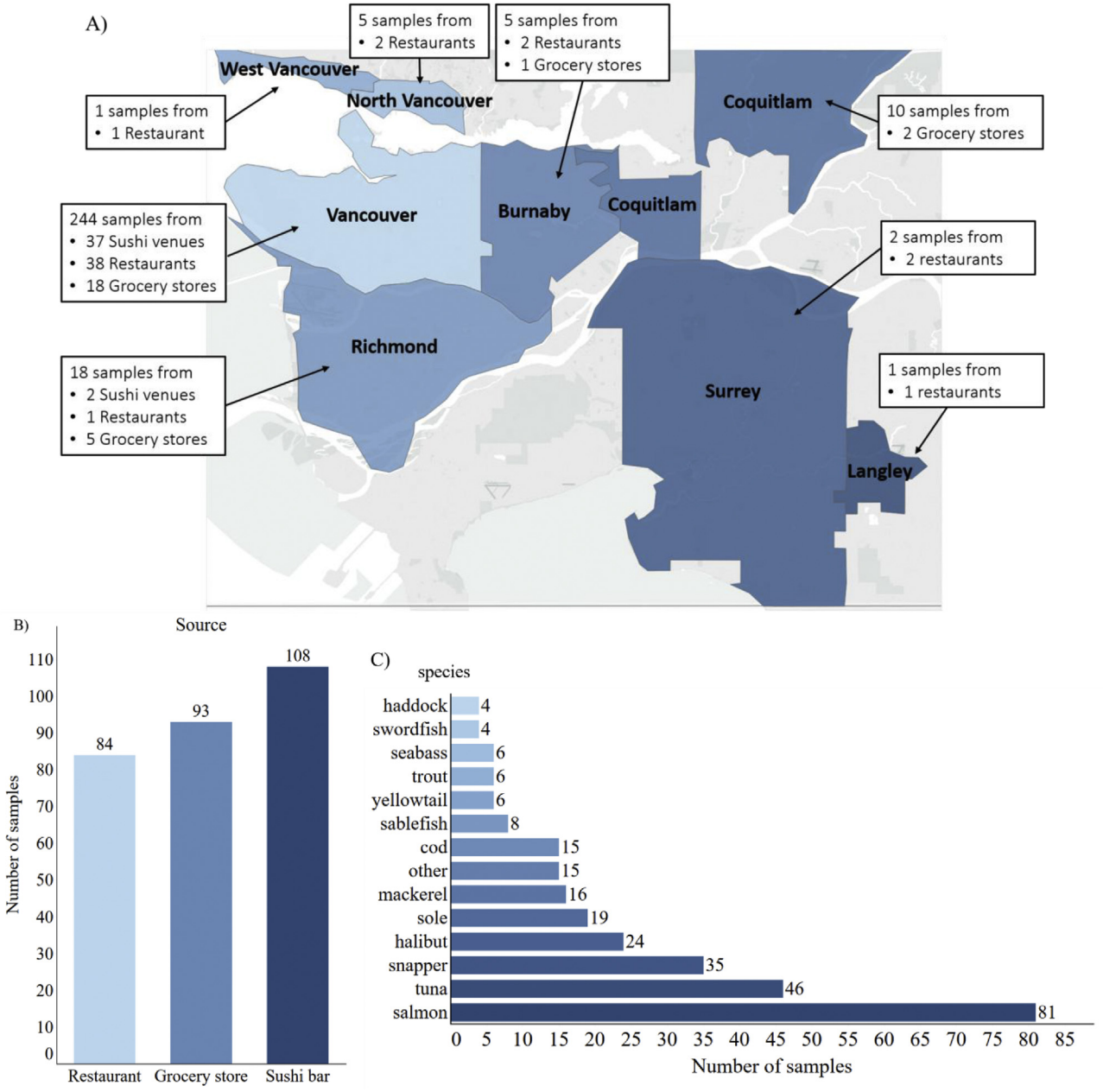

Fig. 1. Categorization of samples collected in Metro Vancouver based on geographical locations (A), source of the samples (B) and major species of samples (C). Snapper in (C) include samples labeled as real snappers (*Lutjanus* spp.) and Pacific snappers (*Sebastes* spp.) and other in (C) include fish species with less than three samples collected as per labeled on the package or menu (*i.e.* amberjack, bigeye, char, herring, monkfish, pollock, pompano, sardine, scad, toothfish, basa, lingcod, seabream).

could be the confusion between snapper (Tai) and genuine snapper (Madai) in sushi culture. Tai and Madai are known as snapper/red snapper and genuine snapper (silver seabream) in sushi culture, respectively, although both Tai and Madai are not acceptable common names in Canada for those fish species (Canadian Food Inspection Agency, 2017a). Besides, two tuna, one seabream and five yellowtail samples were mislabeled. Although Yellowtail is a common vernacular for *Seriola* spp, it is not the approved market name in either Canada or the US. The case of bigeye tuna substituted by Bluefin tuna might indicate a by-catch or IUU fishing issue; while labeling escolar as white tuna is an evident food fraud incident with potential health detriment to susceptible consumers (Unicomb, Kirk, Yohannes, Dalton, & Halliday, 2002) since escolar contains high amounts of indigestible wax esters called gemplylotoxin, a strong purgative that can lead to diarrhea, cramps, nausea or headache (US Food and Drug Administration, 2017c). Mislabeling of seabream was also suspected to be an unintentional practice since there is a high possibility that the owner of the sushi bar failed to recognize the necessity of the term “silver” and the importance of following the CFIA fish list strictly. According to CFIA, yellowtail is the name for a flounder species (*i.e. Limanda ferruginea*), while the biological species of the Yellowtail samples were Japanese amberjack (*Seriola quinqueradiata*) and greater amberjack (*S. dumerili*). Mislabeling involving the misuse of the name amberjack and Yellowtail has been reported by Willette and others (Willette et al., 2017) and is primarily because of the discrepancies in cultures between Japan and Canada and the common use of the term Yellowtail and Hamachi for *Seriola* spp.. Yellowtail indeed refers to the *Seriola quinqueradiata* and *S. dumerili* in Japan, while amberjack is the only acceptable common name for these species in Canada.

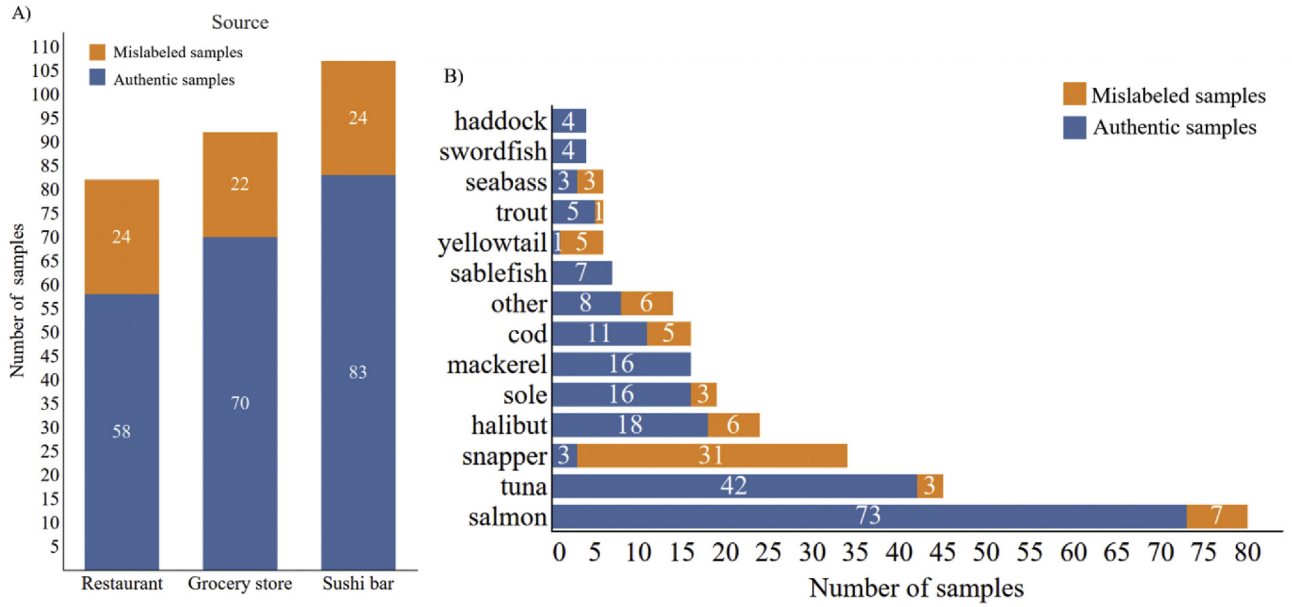

Fig. 2. Number of mislabeled products analyzed based on source of samples (A) and the major species of samples (B). Snapper in (C) include samples labeled as real snappers (*Lutjanus* spp.) and Pacific snappers (*Sebastes* spp.) and other in (C) include fish species with less than three samples collected as per labeled on the package or menu (*i.e.* amberjack, bigeye, char, herring, monkfish, pollock, pompano, sardine, scad, toothfish, basa, lingcod, seabream).

3.3. Non-sushi restaurants

A total of 82 samples purchased from 47 non-sushi restaurants were successfully sequenced with a success rate of 98%. A mislabeling rate of 28% was identified, ranking this as the highest among the three sources (Fig. 2). Again, all the snappers were mislabeled with inappropriate names according to CFIA. Compared with sushi bars, the species of substitution for snappers in non-sushi restaurants were more diverse. Replacement of red snapper by albacore tuna and rockfish (Pacific snappers) as well as snapper by basa was a common fraud. Labeling Pacific snapper as snapper is tied to an ambiguous fish naming system across the Pacific Northwest. Twenty-one species of fish have an accepted market/common name as snapper according to the CFIA fish list, while another eighteen species of fish have a common name including the word “snapper”. However, according to CFIA those eighteen species should not be sold under the “umbrella” term “snapper”. More discussion regarding this is provided in section 3.5.

Mislabeling incidents for salmon, halibut, seabass, cod and sole were also detected. In contrast to the previous studies (Hanner et al., 2011), no Atlantic salmon was sold as Pacific salmon in the restaurant samples that we tested. Substitutions between different Pacific salmons were identified: chum salmon labeled as king and sockeye salmon, as well as sockeye salmon sold as pink salmon. While it is uncertain if the substitution of pink salmon by sockeye salmon indicates an intentional behavior, the substitution of chum salmon for king and sockeye salmon is a highly possible fraud since the price of king and sockeye salmon are around 1.5–2 times that of chum salmon (Wild Alaska Salmon & Seafood Co). A common problem observed in many markets is the naming of steelhead trout. Although *Oncorhynchus mykiss* is an anadramous or ocean-going fish that behaves similarly to salmon, it does not have an accepted common name ‘salmon’ but instead the common name of steelhead salmon or trout. Similar incidents also happened with the misuse of the name cod: black cod (sablefish) being sold as cod in one of our sample. To interpret the motivation of this type of mislabeling practices is challenging since black cod is a very valuable fish species and significantly different from true cod. Possibly the market did not understand the value of adding the term ‘black’ to their marketing and label designation (Xiong, Guardone, et al., 2016). A more understandable incident of mislabeling involved the substitution of cod (3 samples) with the cheaper catfish (*i.e.* basa), an obvious intentional fraud, and one catfish species (*i.e.* striped catfish) has not been included in the CFIA fish list. Another mislabeled cod involves the misidentification of the geographical origin of the fish, which could be an intentional mislabeling practice or an unintentional misidentification due to human error of the staff in the restaurant. Misidentification of geographical origin of halibut was also uncovered in one of our halibut samples (*i.e.* Atlantic halibut labeled as Pacific halibut). Although this type of mislabeling might not damage consumer economic benefit, it could involve selling more vulnerable species to consumers without notification, hindering consumers from making informed decision for wildlife conservation. Besides, greenland turbot (*i.e. Reinhardtius hippoglossoides*) was sold as halibut in two of our samples. Although greenland halibut is a vernacular for *Reinhardtius hippoglossoides* in many countries including Canada, it is not an acceptable market name in Canada, and thus those products should be labeled as turbot. Comparably, Chilean seabass is the vernacular name for Antarctic toothfish in many countries (US Food and Drug Administration, 2017b), but is not an accepted name in Canada. The last two mislabeled halibut samples were substituted with sutchi catfish and haddock, an intentional fraud involving the substitution of a lower value and more readily available species for a high value species. Sutchi catfish, a universally available aquaculture species with a light-color tissue and pleasant taste, were identified as substitutes for a wide variety of high-end fish in our study, including halibut, snapper, sole and cod. The only incident of tuna mislabeling, although the specific species was unresolved using DNA barcoding, might indicate another intentionally fraudulent practice or IUU fishing.

3.4. Grocery stores

A total of 92 samples purchased from 26 grocery stores were sequenced and a success rate of 99% was achieved. A mislabeling rate of 24% was uncovered for fish in grocery stores, ranking the second among the three sampling sources (Fig. 2). Coincidently, snapper and red snapper had the highest mislabeling rate (*i.e.* 73%). The apparent slightly improved labeling situation of snappers in grocery store could be attributed to the acknowledged lack of specificity of DNA barcoding towards the *Sebastes* genus. Two of the snapper samples were identified

Table 2
Details of mislabeled species for fish collected from sushi bars, restaurants, and grocery stores (NIFL represent a species not on the Canadian Food Inspection Agency fish list). Common names not on CFIA list are from US Food and Drug Administration Fish List. Samples with ID containing only digits were purchased from sushi bars, while samples with ID starting with R and G were purchased from restaurants and grocery stores.

Sample ID	City	Labeled name	DNA identified scientific name	Common name on Canadian Food Inspection Agency list
12	Vancouver	red snapper	*Oreochromis niloticus*	tilapia
20	Vancouver	red snapper	*Oreochromis niloticus*	tilapia
25	Vancouver	bigeye tuna	*Thunnus thynnus* or *T. orientalis*	Atlantic or Pacific bluefin tuna
45	Vancouver	Tai	*Oreochromis niloticus*	tilapia
49	Vancouver	Tai	*Oreochromis mossambicus*	tilapia
56	Vancouver	Tai	*Oreochromis niloticus*	tilapia
60	Vancouver	Tai	*Oreochromis niloticus*	tilapia
67	Vancouver	Tai	*Oreochromis niloticus*	tilapia
69	Vancouver	white tuna	*Lepidocybium flavobrunneum*	snake mackerel/escolar
71	Vancouver	Hamachi (yellowtail)	*Seriola dumerili*	greater amberjack
73	Vancouver	Tai	*Oreochromis niloticus*	tilapia
74	Vancouver	Tai	*Oreochromis niloticus*	tilapia
76	Vancouver	Tai	*Oreochromis niloticus*	tilapia
78	Vancouver	Tai	*Oreochromis niloticus*	tilapia
80	Vancouver	Hamachi (yellowtail)	*Seriola quinqueradiata*	Japanese amberjack
83	Vancouver	Tai	*Oreochromis niloticus*	tilapia
91	Vancouver	snapper	*Oreochromis niloticus*	tilapia
95	Vancouver	Yellowtail	*Seriola quinqeradiata*	Japanese amberjack
96	Vancouver	red snapper	*Oreochromis aureus*	blue tilapia
99	Vancouver	snapper	*Oreochromis niloticus*	tilapia
102	Vancouver	seabream	*Pagrus major*	silver seabream
104	Vancouver	Yellowtail	*Seriola quinqeradiata*	Japanese amberjack
109	Vancouver	red snapper	*Dicentrarchus labrax*	silver seabream
111	Vancouver	Yellowtail	*Seriola quinqeradiata*	Japanese amberjack
R6	Vancouver	king salmon (white)	*Oncorhynchus keta*	chum salmon
R11	Vancouver	snapper	*Sebastes entomelas*	Pacific snapper/wido rockfish
R17	Vancouver	snapper	*Sebastes brevispinis*	Pacific snapper/silvergray rockfish
R20	Surrey	snapper	*Sebastes entomelas*	Pacific snapper/wido rockfish
R21	Surrey	pink salmon	*oncorhynchus nerka*	sockeye salmon
R23	Vancouver	steelhead salmon	*Oncorhynchus mykiss*	steelhead salmon/trout
R25	Vancouver	halibut	*Reinhardtius hippoglossoides*	greenland turbot
R30	Vancouver	red snapper	*Thunnus alalunga*	albacore tuna
R35	North Vancouver	red snapper	*Sebastes entomelas*	Pacific snapper/wido rockfish
R42	Vancouver	Ahi tuna	*Thunnus atlanticus* or *T. maccoyii*	blackfin tuna or Southern bluefin tuna
R44	Vancouver	sea bass	*Dissostichus mawsoni*	Antarctic toothfish
R45	Vancouver	cod	*Pangasius hypophthalmus*	sutchi catfish
R46	Burnaby	cod	*Anoplopoma fimbria*	black cod/sablefish
R47	Burnaby	sole	*Pangasianodon hypophthalmus*	sutchi catfish
R49	Vancouver	snapper	*Pangasius hypophthalmus*	sutchi catfish
R50	Burnaby	cod	*Pangasius hypophthalmus*	sutchi catfish
R52	Richmond	halibut	*Reinhardtius hippoglossoides*	greenland turbot
R53	Vancouver	halibut	*Pangasianodon hypophthalmus*	sutchi catfish
R75	Vancouver	Atlantic halibut	*Hippoglossus stenolepis*	Pacific halibut
R76	Vancouver	sockeye salmon	*Oncorhynchus keta*	chum salmon
R79	Vancouver	Pacific halibut	*Melanogrammus aeglefinus*	haddock
R80	Vancouver	Pacific cod	*Gadus morhua*	Atlantic cod
R83	Vancouver	snapper	*Sebastes mystinus* or *S. entomelas*	blue rockfish or widow rockfish
R85	Vancouver	cod	*Pangasianodon hypophthalmus*	NIFL (striped catfish)
G24	Vancouver	Pacific salmon	*oncorhynchus gorbuscha*	pink salmon
G30	Coquitlam	sockeye salmon	*Oncorhynchus gorbuscha*	pink salmon
G41	Coquitlam	sole	*Lepidopsetta polyxystra*	NIFL (Northern rock sole)
G42	Vancouver	sole	*Lepidopsetta polyxystra*	NIFL (Northern rock sole)
G47	Richmond	red snapper	*Lutjanus erythropterus* or *L. malabaricus*	NIFL (crimson snapper) or malabar snapper
G58	Vancouver	Snapper	*Sebastes entomelas*	Pacific snapper/wido rockfish
G65	Vancouver	Chilean seabass	*Dissostichus eleginoides*	Patagonian toothfish
G67	Vancouver	snapper	*Sebastes alutus*	redfish/rockfish/Pacific Ocean perch
G71	Vancouver	salmon	*Oncorhynchus nerka*	sockeye salmon
G74	Richmond	basa	*Oreochromis niloticus*	tilapia
G78	Vancouver	snapper	*Sebastes alutus*	redfish/rockfish/Pacific Ocean perch
G82	Vancouver	snapper	*Sebastes entomelas*	Pacific snapper/wido rockfish
G86	Vancouver	yellowtail scad	*Atule mate*	NIFL (yellowtail scad)
G87	Vancouver	red big eye	*Priacanthus tayenus*	NIFL (purple spotted bigeye)
G90	Richmond	menhaden	*Clupea pallasii*	Pacific herring
G91	Richmond	wild salmon	*Salmo salar*	Atlantic salmon
G93	Vancouver	golden pompano	*Trachinotus ovatus*	NIFL (pompano)
G119	Vancouver	Chilean seabass	*Dissostichus eleginoides*	Patagonian toothfish
G120	Vancouver	Pacific snapper	*Sebastes alutus*	Pacific Ocean perch
G122	Vancouver	Atlantic halibut	*Hippoglossus stenolepis*	Pacific halibut
G124	Vancouver	snapper	*Sebastes zacentrus*	sharpchin rockfish
G129	Vancouver	snapper	*Sebastes brevispinis*	Pacific snapper/silvergray rockfish

Table 3
Results of generalized linear regression model with binary distribution for the analysis of properties related to fishes having significant impact on the mislabeling/fraud practices ($\alpha = 0.05$).

	F-stat	DF1	DF2	p-Value
intercept	4.69	1	271	0.021
source	0.52	2	271	0.59
raw/cooked	1.64	1	271	0.22
appearance	3.08	2	271	0.061
product form	3.45	3	271	0.020
color of the flesh	20.70	1	291	2.62×10^{-5}

as *Sebastes mystinus* or *S. entomerlas* with the same veracity. Although *S. mystinus* is not a species being included in CFIA Fish List, samples were correctly labeled if they are *S. entomerlas.* Therefore, these two samples were identified as properly labelled samples in this current study. For the mislabeled snapper samples, although the species substitution in grocery stores are all in the same genus as Pacific snappers (*Sebastes* spp.) and real snappers (*Lutjanus* spp.) and they all possess a name with the term "snapper", these fish should be labelled appropriately according to the CFIA fish list using their specific names instead of being labeled with an "umbrella" term snapper.

Atlantic salmon sold as wild salmon and pink salmon labeled as sockeye salmon might represent food fraud incidents, while the other two mislabeled salmon products are due to the noncompliance with the "Labelling of Pacific Salmon in Canada" requirement (Canadian Food Inspection Agency, 2017c). The specific species of Pacific salmon should be clearly listed on the label. The misuse of the term Chilean seabass for Patagonian toothfish were also identified in two grocery store samples. Mislabeling of tilapia as basa and Pacific herring as menhaden was detected. However, the motivations behind these were difficult to understand. Although menhaden is not a name included in CFIA fish list, this common name is used for *Ethmidium maculatum* in the US, a species of fish belonging to the same family as herring (*i.e. Clupeidae*). In addition, *Ethmidium maculatum* does not possess an acceptable English name or French name in the CFIA fish list, posing a gap in labeling standards. Therefore, this mislabeling incident might be an unintentional behavior. Similar to the case of restaurant samples, misidentification of geographical origin of one halibut sample was also detected in the grocery sample, although this time a less concerned species (Pacific halibut) was sold as Atlantic halibut which was endangered according to International Union for Conservation of Nature (IUCN) Red List (International Union for Conservation of Nature, 2017).

Beside substituting a product with another fish included in the CFIA fish list, there are also several samples collected from grocery stores that were identified as species outside the list, including *Trachinotus ovatus*, *Priacanthus tayenus*, *Lepidopsetta polyxystra, Atule mate,* and *Sebastes serranoides*. Although *Priacanthus* spp., *Trachinotus* spp. and *Sebastes* spp. are in the CFIA fish list, general common names of bigeye, pompano and rockfish are assigned. According to the FDA fish list, only *Priacanthus macracanthus* can be labeled as red bigeye while no fish in CFIA has a common name of golden pompano. The genus *Atule* and species *Lepidopsetta polyxystra* are not on the CFIA fish list, while according to the FDA fish list these samples were labeled correctly. Therefore, these mislabeling incidents demonstrate some gaps in the CFIA fish list and in harmonization between countries.

3.5. Ambiguity of labeling regulation

Apart from several obvious economically motivated mislabeled products (*e.g.* tilapia and albacore tuna for red snapper and snapper, chum salmon for king salmon, and basa for cod, sole and halibut), other mislabeling incidents tend to be associated with complicated fish naming systems and non-harmonized fish labeling requirements among countries.

Snappers are one of the fish groups having a notorious naming system. Strictly speaking, snappers refer to 112 species in 17 genera of the Lutjanidae family (Cawthorn & Mariani, 2017), primarily harvested in tropical and sub-tropical areas. Nevertheless, "snapper" has been used as the vernacular name, and thus to some extent accepted by local regulatory agencies, for many species outside the family of Lutjanidae. For example, snappers commonly refer to seabream in the Sparidae family in New Zealand (Cawthorn & Mariani, 2017) and include rockfish species in *Sebastes* and *Sebastolobus* genus of Scorpaenidae family in Canada (Canadian Food Inspection Agency, 2017a). Among the mislabeled snapper and red snapper samples tested in this current study, 15 out of 23 samples were found as rockfish in the *Sebastes* genus. In Canadian fish markets, there are 14 rockfish species with a common name containing "snapper" in Scorpaenidae family. Only one of them can be labeled as snapper or red snapper (*Sebastes ruberrimus*), while the others are accepted as Pacific snapper. Therefore, it is challenging to determine whether such mislabeling is intentional or merely because of a confusing use of a common name. Noting that real red snapper (*Lutjanus campechanus*) and some snappers in the Lutjanidae family are depleted due to overfishing, the status of some rockfish (Pacific snapper) species is also not healthy. Although the Ocean Wise program managed by the Vancouver Aquarium has updated their assessment on ground fish stocks and recommended four species of rockfish (*i.e.* yellowtail, yellowmouth, silvergray, and canary rockfish) caught by mid-water trawl as sustainable sources (Brown, 2016), only one of the substitutions identified in our study belong to those four species. Consequently, labeling rockfish as snappers could not only cause overestimation of real snapper stock status and impact consumers' perception on this group of fish, but also open the door for replacing fish with another species that has conservation concerns. To avoid such type of mislabeling, requiring greater detail on the type of fish, including the specific scientific name on the label or menu could be effective.

Mislabeling induced by discrepancies in the vernacular among countries is also ubiquitous. Labeling amberjack as yellowtail in sushi restaurants was identified in our study and has been reported previously (Willette et al., 2017). In the context of sushi, Yellowtail (*i.e.* Hamachi) refers to amberjack. However, in Canada, yellowtail refers to yellowtail flounder. To best maintain the authenticity of the sushi culture but to not mislead consumers about the identity of a fish, updating the CFIA fish list to include Yellowtail as an accepted common name for amberjack or require the addition of specific scientific name is recommended. Inconsistency in common names also resulted in the mislabeling of Patagonian and Antarctic toothfish.

For the five samples that were species not included in the CFIA fish list, three of them were correctly labeled according to the FDA fish list, while two were labeled correctly with the generic name. The US, as the major importing and exporting country of Canadian seafood, has 1880 species included in their fish list (US Food and Drug Administration, 2017b), while the Canadian fish list contains only 919 individual Taxonomic Serial Numbers (Canadian Food Inspection Agency, 2017a). Although some species are covered by the genus name (*e.g. Priacanthus* spp.), there are genus that are accepted by the US but not by Canada (*e.g. Atule*), potentially hindering smooth and transparent trading between these two countries. Moreover, the accepted common names for many fish species are not uniform between countries, rendering problems in assigning the correct labels to be approved by the customs in each country (Cawthorn & Mariani, 2017). This scenario is even worse when trading between countries with different official languages. To tackle such problems, harmonizing the accepted common names between trading countries and requiring the use of scientific names on label would provide a solution.

Labeling products with scientific name not only aids in ensuring the precision of labeling, but also contributes to the conservation of vulnerable species. Fourteen species and 15 species of fish in the Scombridae family can be collectively labeled as tuna in Canada and US, respectively. However, they differ significantly in their stock status,

vulnerability to overfishing, market values, and nutritional values. According to the IUCN Red List (International Union for Conservation of Nature, 2017), Southern bluefin tuna (*Thunnus maccoyii*) is critically endangered, followed by Atlantic bluefin tuna being endangered and bigeye tuna and Pacific bluefin tuna being vulnerable, while albacore and yellowfin tuna are near threatened. Lumping all the tuna species under the single "umbrella" term of tuna could create loopholes for IUU fishing of species with unhealthy stock, pose challenges in stock status evaluation, and diminish the effectiveness of fishery management. Consumer preference for tuna species varies significantly and thus impacts the market price for the different tuna species, with bluefin tunas being the most favored species and the most expensive ones. Furthermore, various tuna species differ in their level of heavy metal content and there are consumption advisories for some tunas for susceptible populations (*e.g.* pregnant women and children). According to FDA, fresh/frozen bigeye tuna have the highest mercury level, followed by fresh/frozen albacore and yellowfin tuna and canned albacore tuna, while fresh/frozen and canned skipjack tuna had the lowest mercury level (US Food and Drug Administration, 2014). Another study conducted by Lowenstein and others (Lowenstein et al., 2010) concluded that bluefin tuna had a similar or even higher level of mercury compared with bigeye tuna and suggested the consumption of bluefin tuna should also be avoided by certain vulnerable populations (Lowenstein et al., 2010). Consequently, using a collective name for the different tuna species could impede people from making informed decisions based on their dietary restrictions and conservation risks of fish. Similarly, in the case of halibut, use of the scientific name or regional classification for Pacific halibut and Atlantic halibut would be helpful. Requiring the unique scientific name could make the protection of Atlantic halibut more effective.

Another factor associated with the little improvement in the fish market integrity in Vancouver or Canada is lack of adequate traceability. According to CFIA, labeling the country of origin for imported products where the last transformation took place is compulsory (Canadian Food Inspection Agency, 2017c). However, providing the geographic location where the fish was harvested or produced is voluntary. Having complex and fragmented fish supply and production chains that consist of transit and processing in several different countries makes regulatory compliance with the labeling requirements in the country to which the fish is destined, difficult. Labeling a product without sufficient information on the processing and/or trading histories can significantly increase challenges in tracing and tracking of fish for either IUU purposes or for species identification. For example, for a fish caught in Canada, shipped to China for gutting and filleting, breaded in the US and sold in Canada should be labeled as a product of the US. However, every single node in the supply chain offers opportunities for mislabeling and fraudulent practices. To ensure the integrity of products which have traveled around the world, detailed information on processing and trading histories is mandatory. Labeling fish products with information of their geographical origins is of great significance, because harvested or produced at different locations, the same fish species could have significant difference in nutritional values, level of contaminations, and/or conservation risk levels (Sunderland, 2007). In addition, labeling the product with catching methods provides another dimension for understanding and tracing the fish back to the source and thus can help eliminate IUU fishing products from appearing in fish market in Canada, although this does not mean that the IUU fish would not be present in some other markets.

4. Conclusions

The identity of 281 fish products purchased from sushi bars, non-sushi restaurants and grocery stores in Metro Vancouver area was successfully analyzed using DNA barcoding and DNA mini-barcoding methods. An average mislabeling rate of 25% was observed for products collected from the three sources, which was comparable to the mislabeling level in 2010. Mislabeling rate in sushi bars, non-sushi restaurants and grocery stores were 22%, 28%, and 24%, respectively. Snappers were the fish group with the highest mislabeling rate. Based on the type of substitution found, both intentional fraud, unintentional misidentification and/or unintentional misuse of vernacular names were observed. Irrespective of motivations, mislabeling of fish products could jeopardize consumer's health, damage the economic interest of consumers as well as the entire fishing industry, and weaken the effectiveness of fish management and conservation measures. To improve the integrity of the fish market in Canada, several actions are recommended. First, the CFIA fish list should be harmonized with major trading countries to facilitate trade and avoid mislabeling. Secondly, mandatory labeling with scientific name can reduce the opportunity of mislabeling associated with historical use of confusing and ambiguous vernacular names and mistranslation. Replacing vague generic names to cover multiple species with notable differences in fishing pressure and stock health, nutrition and contamination levels, and market values should be avoided, again by requiring the use of scientific names across the value chain. Last but not the least, including the information of where the fish was farmed or caught, the processing history and the fishing/farming methods used enables the effective boat-to-table tracing and tracking of fish products and reduces the potential for fraud.

Compete of interest

The authors declare no conflict of interest.

Acknowledgement

This study is funded by Peter Wall Institute for Advanced Studies awarded to Dr. Xiaonan Lu. The authors sincerely thank Dr. Barbara Rasco and Joan Hewer for providing suggestions and comments and helping to polish the writing.

Appendix A. Supplementary data

Supplementary data related to this article can be found at http://dx.doi.org/10.1016/j.foodcont.2018.06.023.

References

Agriculture and Agri-Food Canada (2016). Industry overview for fish and seafood. Retrieved Jan 24 2018, from http://www.agr.gc.ca/eng/industry-markets-and-trade/market-information-by-sector/fish-and-seafood/industry-overview/?id=1383756439917.

Armani, A., Guardone, L., La Castellana, R., Gianfaldoni, D., Guidi, A., & Castigliego, L. (2015). DNA barcoding reveals commercial and health issues in ethnic seafood sold on the Italian market. *Food Control, 55*, 206–214. https://doi.org/10.1016/j.foodcont.2015.02.030.

Brown, A. (2016). Select B.C. rockfish now recommended by Vancouver aquarium's Ocean Wise program. Retrieved Feb 3 2018, from https://ocean.org/media-releases/select-b-c-rockfish-now-recommended-vancouver-aquariums-ocean-wise-program/.

Canadian Food Inspection Agency (2013). *Regulatory verification system of the fish inspection program fish inspection program sampling policy.* Canadian Food Inspection Agency.

Canadian Food Inspection Agency (2017a). *CFIA fish list.* Canadian Food Inspection Agency. Retrieved Feb 5, 2018, from http://www.inspection.gc.ca/active/scripts/fssa/fispoi/fplist/fplist.asp?lang=e.

Canadian Food Inspection Agency (2017b). *Guidance on determining the common names for fish sold or processed in Canada.* Canadian Food Inspection Agency. Retrieved Mar 1 2018, from http://www.inspection.gc.ca/food/fish-and-seafood/product-inspection/fish-list/common-names/eng/1352987508427/1352993955238.

Canadian Food Inspection Agency (2017c). Labelling requirements for fish and fish products. Generic common names. Retrieved Feb 2 2018, from http://www.inspection.gc.ca/food/labelling/food-labelling-for-industry/fish-and-fish-products/eng/1393709636463/1393709677546?chap=0-s2c2.

Cawthorn, D.-M., & Mariani, S. (2017). Global trade statistics lack granularity to inform traceability and management of diverse and high-value fishes. *Scientific Reports, 7*(1), 12852. http://dx.doi.org/10.1038/s41598-017-12301-x.

Cawthorn, D.-M., Steinman, H. A., & Witthuhn, R. C. (2012). DNA barcoding reveals a high incidence of fish species misrepresentation and substitution on the South African market. *Food Research International, 46*(1), 30–40. https://doi.org/10.1016/j.foodres.2011.11.011.

Chin Chin, T., Adibah, A. B., Danial Hariz, Z. A., & Siti Azizah, M. N. (2016). Detection of mislabelled seafood products in Malaysia by DNA barcoding: Improving transparency in food market. *Food Control, 64*, 247–256. https://doi.org/10.1016/j.foodcont.2015.11.042.

Cline, E. (2012). Marketplace substitution of Atlantic salmon for Pacific salmon in Washington State detected by DNA barcoding. *Food Research International, 45*(1), 388–393. https://doi.org/10.1016/j.foodres.2011.10.043.

Council of the European Union & European Parliament (2013). Regulation (EU) No 1379/2013 of the European parliament and of the council of 11 december 2013 on the common organisation of the markets in fishery and aquaculture products, amending council regulations (EC) No 1184/2006 and (EC) No 1224/2009 and repealing council regulation (EC) No 104/2000. *Official Journal of the European Union, L, 354*, 21.

Fisheries and Oceans Canada (2012). *Canadian fisheries statistics 2011-12.* Retrieved from.

Food and Agriculture Organization (2016). *The state of world fisheries and aquaculture.* Retrieved from Rome.

Giusti, A., Armani, A., & Sotelo, C. G. (2017). Advances in the analysis of complex food matrices: Species identification in surimi-based products using Next Generation Sequencing technologies. *PLoS One, 12*(10), e0185586. http://dx.doi.org/10.1371/journal.pone.0185586.

Hajibabaei, M., Smith, M. A., Janzen, D. H., Rodriguez, J. J., Whitfield, J. B., & Hebert, P. D. N. (2006). A minimalist barcode can identify a specimen whose DNA is degraded. *Molecular Ecology Notes, 6*(4), 959–964. http://dx.doi.org/10.1111/j.1471-8286.2006.01470.x.

Handy, S. M., Deeds, J. R., Ivanova, N. V., Hebert, P. D. N., Hanner, R., Ormos, A., ... Yancy, H. F. (2011). Single laboratory validated method for DNA-barcoding for the species identification of fish. Retrieved from https://www.fda.gov/Food/FoodScienceResearch/DNASeafoodIdentification/ucm237391.htm.

Hanner, R., Becker, S., Ivanova, N. V., & Steinke, D. (2011). FISH-BOL and seafood identification: Geographically dispersed case studies reveal systemic market substitution across Canada. *Mitochondrial DNA, 22*(sup1), 106–122.

International Union for Conservation of Nature (2017). The IUCN red list of threatened species. From international union for conservation of nature. http://www.iucnredlist.org/about/citing.

Jacquet, J. L., & Pauly, D. (2008). Trade secrets: Renaming and mislabeling of seafood. *Marine Policy, 32*(3), 309–318. https://doi.org/10.1016/j.marpol.2007.06.007.

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 Control, 56*, 71–76. https://doi.org/10.1016/j.foodcont.2015.03.007.

Levin, J. (2017). Seafood fraud and mislabelling in ottawa. Retrieved from http://www.oceana.ca/en/publications/reports/seafood-fraud-and-mislabelling-ottawa.

López-Andreo, M., Aldeguer, M., Guillén, I., Gabaldón, J. A., & Puyet, A. (2012). Detection and quantification of meat species by qPCR in heat-processed food containing highly fragmented DNA. *Food Chemistry, 134*(1), 518–523. https://doi.org/10.1016/j.foodchem.2012.02.111.

Lowenstein, J. H., Burger, J., Jeitner, C. W., Amato, G., Kolokotronis, S.-O., & Gochfeld, M. (2010). DNA barcodes reveal species-specific mercury levels in tuna sushi that pose a health risk to consumers. *Biological Letters, 6*(5), 692–695. http://dx.doi.org/10.1098/rsbl.2010.0156.

Mariani, S., Ellis, J., O'Reilly, A., Bréchon, A. L., Sacchi, C., & Miller, D. D. (2014). Mass media influence and the regulation of illegal practices in the seafood market. *Conserv Lett, 7*(5), 478–483. http://dx.doi.org/10.1111/conl.12085.

Mariani, S., Griffiths, A. M., Velasco, A., Kappel, K., Jérôme, M., Perez-Martin, R. I., ... Sotelo, C. G. (2015). Low mislabeling rates indicate marked improvements in European seafood market operations. *Frontiers in Ecology and the Environment, 13*(10), 536–540. http://dx.doi.org/10.1890/150119.

McGill University (2010). *Sanger sequencing user guide.* McGill University and Genome Quebec Innovation Centre.

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 Anal Method, 9*(10), 2711–2720. http://dx.doi.org/10.1007/s12161-016-0460-3.

Naaum Amanda, M., & Hanner, R. (2015). Community engagement in seafood identification using DNA barcoding reveals market substitution in Canadian seafood. *DNA Barcodes, 3*(1), 74. http://dx.doi.org/10.1515/dna-2015-0009.

Naaum, A. M., Warner, K., Mariani, S., Hanner, R. H., & Carolin, C. D. (2016). *Chapter 1-seafood mislabeling incidence and impacts seafood authenticity and traceability.* San Diego: Academic Press3–26.

Oceana (2015). Too cheap to be true: Seafood fraud in Brussels. Retrieved from http://eu.oceana.org/sites/default/files/421/oceana_factsheet_seafood_fraud_brussels_eng.pdf.

Oceana Canada (2017). Mystery fish. Retrieved from http://www.oceana.ca/en/publications/reports/mystery-fish-seafood-fraud-canada-and-how-stop-it.

Reilly, A. (2018). Overview of food fraud in the fisheries sector. Retrieved from http://www.fao.org/documents/card/en/c/I8791EN/.

Shokralla, S., Hellberg, R. S., Handy, S. M., King, I., & Hajibabaei, M. (2015). A DNA mini-barcoding system for authentication of processed fish products. *Scientific Reports, 5*, 15894. http://dx.doi.org/10.1038/srep15894https://www.nature.com/articles/srep15894-supplementary-information.

Sunderland, E. M. (2007). Mercury exposure from domestic and imported estuarine and marine fish in the U.S. seafood market. *Environmental Health Perspectives, 115*(2), 235–242. http://dx.doi.org/10.1289/ehp.9377.

Tinacci, L., Stratev, D., Vashin, I., Chiavaccini, I., Susini, F., Guidi, A., et al. (2018). Seafood labelling compliance with European legislation and species identification by DNA barcoding: A first survey on the Bulgarian market. *Food Control, 90*, 180–188. https://doi.org/10.1016/j.foodcont.2018.03.007.

Unicomb, L., Kirk, M., Yohannes, K., Dalton, C., & Halliday, L. (2002). An outbreak of gastrointestinal illness associated with the consumption of escolar fish. *Commun Dis Intell Q Rep, 26*(3), 441–445.

US Food and Drug Administration (2013). *Focused mitigation strategies to protect food against intentional adulteration.* Retrieved fromhttps://www.regulations.gov/docket?D=FDA-2013-N-1425.

US Food and Drug Administration (2014). Mercury levels in commercial fish and shellfish (1990-2012). Retrieved Feb 4 2018, from https://www.fda.gov/food/foodborneillnesscontaminants/metals/ucm115644.htm.

US Food and Drug Administration (2017a). Protecting the food supply from intentional adulteration, such as acts of terrorism. Retrieved Jan24 2018, from https://www.fda.gov/Food/GuidanceRegulation/FSMA/ucm587803.htm.

US Food and Drug Administration (2017b). The seafood list. From US food & Drug administtration. https://www.accessdata.fda.gov/scripts/fdcc/?set=seafoodlist.

US Food and Drug Administration (2017c). US FDA fish and fishery products hazard and controls guidance. Retrieved Mar 2 2018, from https://www.fda.gov/Food/GuidanceRegulation/GuidanceDocumentsRegulatoryInformation/Seafood/ucm2018426.htm.

Vandamme, S. G., Griffiths, A. M., Taylor, S.-A., Di Muri, C., Hankard, E. A., Towne, J. A., ... Mariani, S. (2016). Sushi barcoding in the UK: Another kettle of fish. *PeerJ, 4*, e1891. http://dx.doi.org/10.7717/peerj.1891.

Wang, C.-s., Van Fleet, D. D., & Mishra, A. K. (2017). Food integrity: A market-based solution. *British Food Journal, 119*(1), 7–19. http://dx.doi.org/10.1108/BFJ-04-2016-0144.

Ward, R. D., Hanner, R., & Hebert, P. D. N. (2009). The campaign to DNA barcode all fishes, FISH-BOL. *Journal of Fish Biology, 74*(2), 329–356. http://dx.doi.org/10.1111/j.1095-8649.2008.02080.x.

Warner, K., Mustain, P., Carolin, C. D., Disla, C., Kroner, R. G., Lowell, B., et al. (2015). Oceana reveals mislabeling of America's favorite fish: Salmon. Retrieved from https://usa.oceana.org/sites/default/files/salmon_testing_report_finalupdated.pdf.

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.

Wild Alaska Salmon & Seafood Co. The difference between salmon species. Retrieved Feb 1 2018, from https://wildalaskasalmonandseafood.com/the-difference-between-salmon-species/.

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. *Conservation Biology, 31*(5), 1076–1085. http://dx.doi.org/10.1111/cobi.12888.

Wong, E. H. K., & Hanner, R. H. (2008). DNA barcoding detects market substitution in North American seafood. *Food Research International, 41*(8), 828–837. https://doi.org/10.1016/j.foodres.2008.07.005.

Xiong, X., D'Amico, P., Guardone, L., Castigliego, L., Guidi, A., Gianfaldoni, D., et al. (2016a). The uncertainty of seafood labeling in China: A case study on cod, salmon and tuna. *Marine Policy, 68*, 123–135. https://doi.org/10.1016/j.marpol.2016.02.024.

Xiong, X., Guardone, L., Giusti, A., Castigliego, L., Gianfaldoni, D., Guidi, A., et al. (2016b). DNA barcoding reveals chaotic labeling and misrepresentation of cod (鳕, Xue) products sold on the Chinese market. *Food Control, 60*, 519–532. https://doi.org/10.1016/j.foodcont.2015.08.028.