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“HORSE MEAT MARKET SURVEY AND DNA TESTING”

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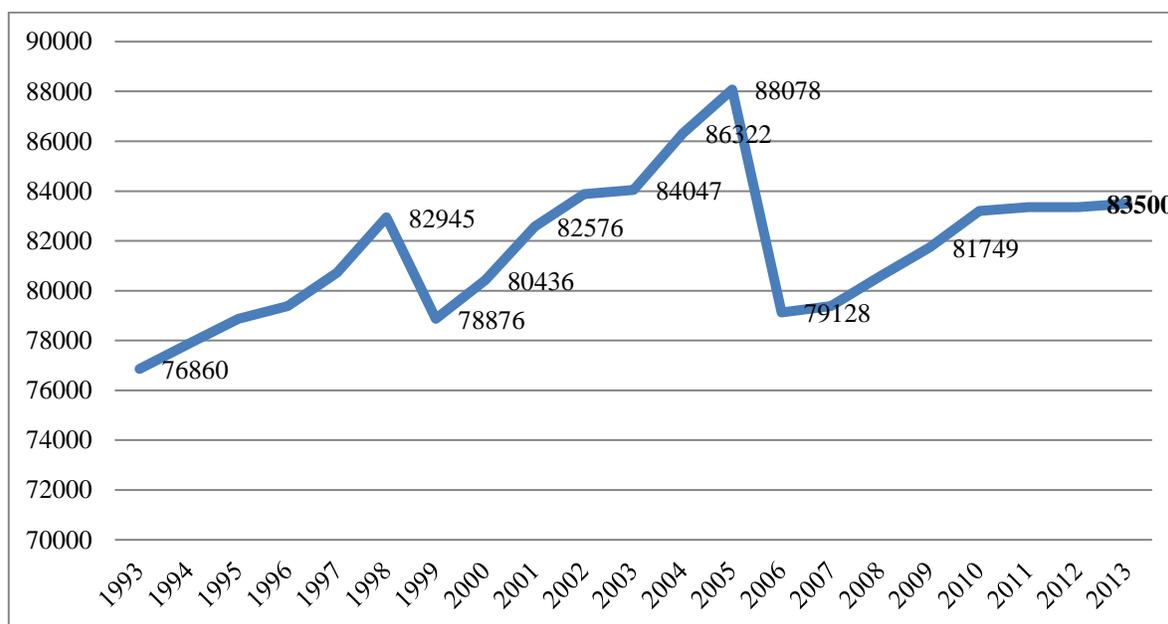
EXECUTIVE SUMMARY

The objective of the project was to carry out a brief market survey of horsemeat and a blind horse-DNA testing in “meat” in Mexico. The experimental design included six Mexican cities (Aguascalientes, Zacatecas, Chihuahua, Mexico City, Pachuca, and San Vicente Chicoloapan), five different market places (butcher shops, supermarkets, permanent street markets, mobile street markets, and others), and four types of meat samples (ground meat, regular *tacos*, crispy *tacos*, and thin steaks [*bistec*]) unlabeled or labeled as beef. Researchers in this study carried out Study 1 (Market survey of horsemeat sold in Mexico) and Study 2 (DNA testing to detect horsemeat in generic meat sold as beef) from January to June of 2016. The total number of samples analyzed was 433. For Study 1, we conducted 339 surveys and results indicated that the majority (>80%) of industry-related personnel do not wish to sell horsemeat and they do not know any horsemeat suppliers. The research team observed poor sanitary conditions in most outlets, with the exception of supermarkets. In Study 2, results from DNA testing showed that 9.93% of the samples of products sold in the different outlets were positive for horsemeat (43 samples). Mexico City, Chihuahua, and Pachuca had around 12% of positive samples, while Aguascalientes had 11% and Zacatecas had 7%. Ground meat is the sample type with the highest percentage of samples positive for horsemeat (42%). The 9.93% found could be due to the fact we directed the investigations to cities with official slaughterhouses processing horses and poorly regulated markets. We carried out a complementary analysis on clenbuterol presence in 29 of the samples that tested positive for horsemeat (only raw meat could be tested; cooked meat could not be tested). We found significant clenbuterol concentrations in 93.10% of the 29 tested samples.

I. BACKGROUND

Mexico is the second largest horsemeat producer in the world, second only to China, and a major exporter of this product. The Food and Agriculture Organization of the United Nations (FAO) defines “meat” as “the flesh of animals used for food”. From this standpoint, according to the same source, 83,500 tonnes of horse flesh were produced in Mexico in 2013 (Figure A).¹ As for the official records of the Mexican Ministry of Trade, in 2015 Mexico exported 2,927,933 kilograms of horsemeat worth 8,924,758 U.S. dollars, mainly to Vietnam and Russia.²

Figure A. Volume of horsemeat production in Mexico, 1961-2013 (tonnes).



Source: FAOSTAT database, “Production”, accessed 20 October, 2016.

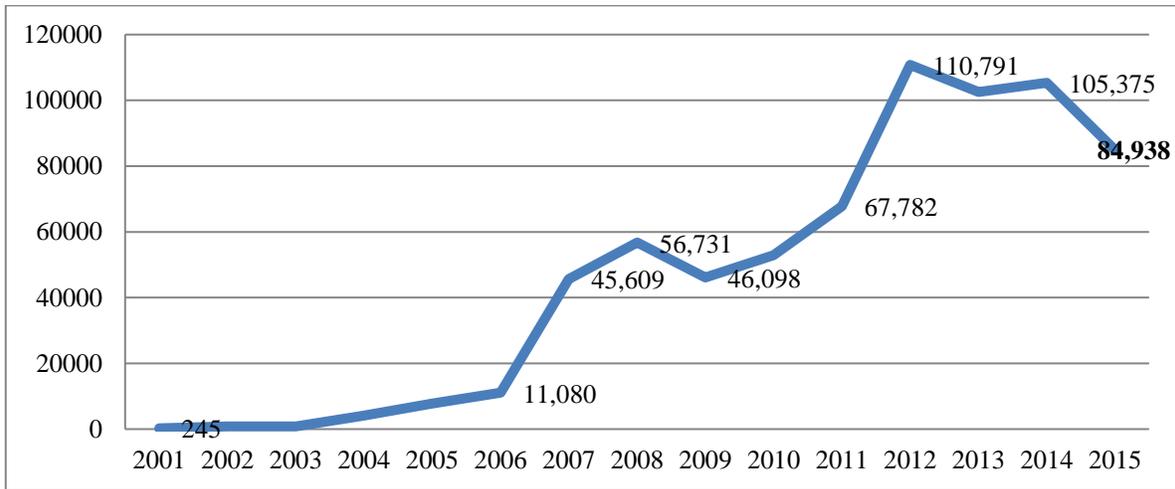
Horsemeat production in Mexico has augmented in the last decade largely as a result of the increase in the number of U.S. horses exported to the southern country for slaughter (Figure B). According to U.S. Department of Agriculture (USDA), in 2006, one year before

¹ The database does not differentiate between horsemeat production intended for human consumption, which may be destined to feed animals. However, it gives an overview of the magnitude of horse slaughter on an overall level. Food and Agriculture Organization of the United Nations, FAOSTAT Statistics Database, “Livestock Primary Meat Horse”, <http://faostat3.fao.org/browse/O/OL/E> [Accessed 25 May, 2016]

² Secretaría de Economía, *SIAMI Sistema de Información Arancelaria Vía Internet*, <http://www.economia-snci.gob.mx/> [Accessed 28 October, 2016].

that the U.S. slaughter plants were closed, exporters sent more than 11,000 live horses to Mexico; by 2012 the number reached a peak at nearly 111,000. This increase of 1,000% stems from the closure of the last remaining horse slaughterhouses in the U.S. following the withdrawal of funding for USDA slaughter plants inspections.³

Figure B. Number of American Horses Exported for Slaughter to Mexico, 2001-2015.



Source: Annual Data for USDA, *Foreign Agricultural Service*, “FAS Agricultural Import Aggregations and HS-10 Digit Import Commodities (Live Horses for Immediate Slaughter)”, <http://www.fas.usda.gov/data> [Accessed 03 November, 2016]

Until recently, Mexico was one of the main suppliers of horsemeat to the European Union (EU), with Belgium, the Netherlands, and France being the main importing countries.⁴ The European Commission imposed a conditional ban on horsemeat imports from Mexico (where 87% of horses slaughtered for export to the EU were of U.S. origin) after the Food and Veterinary Office’s latest audit concluded that it is not possible to guarantee the reliability of producer’s declarations or the traceability of the horses slaughtered for meat originating from the U.S. and Mexico. The decision of the European Commission also applied to Switzerland on the basis of the Bilateral Veterinary Agreement.⁵

³ The current law does not prohibit the shipment of U.S. horses to neighbouring countries. Tadlock Cowan, “Horse Slaughter Prevention Bills and Issues”, *CRS Report for Congress*, 28 June 2013, 8.

⁴ Secretaría de Economía, *SIAMI Sistema de Información Arancelaria Vía Internet*, <http://www.economia-snci.gob.mx/> [Accessed 28 October, 2016].

⁵ The main concern of the EU authorities is related to the controls on traceability of live horses, which “do not achieve standards comparable to those specified in EU legislation”. European Commission, *Final report of an audit carried out in Mexico from 24 June to 04 July 2014 in order to evaluate the operation of controls*

After the EU suspension came into force in 2015, media outlets in Mexico have published reports of meat mislabeling practices in Aguascalientes, Chihuahua, and other states, where horsemeat would be sold unlabeled or mislabeled.⁶ Scholars have identified and documented meat mislabeling practices in Mexico in the past.⁷

Mexico continues to be a major horsemeat exporter. Nonetheless, the statistics suggest that the product is mainly consumed internally. Regarding the marketing of horse meat in Mexico City, the average price of retail is 70 Mexican pesos versus 140 Mexican pesos for beef meat. Thus, given the profitability of horsemeat *vis-à-vis* beef, distorted practices could have arisen.

In Mexico, as in other countries, horsemeat consumption is often associated with low status, poverty, and ignorance. A poll conducted by *Parametría*, a Mexican polling agency, reveals that the majority of Mexicans are opposed to eating horsemeat and would support measures to ensure that horse meat is not mislabeled on a consistent basis. According to the poll conducted in 2015, 74% of Mexicans claim they have never eaten and would never eat horsemeat, 73% would be very upset if they found out that they had consumed mislabeled horsemeat, and 96% support strong supervision to avoid meat mislabeling in Mexico.⁸

Besides its social stigma, the consumption of horsemeat could pose risks to human health. Horses from the U.S. and Mexico are raised and treated as companion animals, not as food-producing animals.⁹ Unlike animals raised for food, the vast majority of horses sent to

over the production of fresh horse meat and meat products intended for export to the European Union, including monitoring of residues and contaminants as well as certification procedures, DG(SANCO), 2014-7223 – MR FINAL, 2014, p. 7.

⁶ Sara Álvarez F., “Investiga la Profeco la Venta de Carne de Caballo en Carnicerías, *Página 24*, Aguascalientes, 2 March, 2015. José T. Méndez, “Aumenta consumo de carne de caballo”, *AM León*, León, 7 April, 2015.

⁷ Mislabeling prevents customers from making informed food choices for purchase. [D. E. Kane and R. S. Hellberg, “Identification of species in ground meat products sold on the U.S. commercial market using DNA-based methods”, *Food Control*, 59 (2016), 158-163]. A previous case study for Mexico has reported mislabeling rates between 20% and 70% for meat products such as sausages, ground meat, and dried meats [M.E. Flores-Munguía, M.C. Bermudez-Almada and L. Vazquez-Moreno, “A research note: detection of adulteration in processed traditional meat products”, *Journal of Muscle Foods*, 11 (2000), 319-325].

⁸ The poll was conducted between 25 and 29 July, 2015 and 800 Mexicans were surveyed. *Parametría: Investigación estratégica. Carta Paramétrica. Consumo de carne de caballo, 2015.*

⁹ H.G. González, D. Mota-Rojas, M. Becerril-Herrera, C. Casas-García, M. Zermeño, R. Ramírez-Necochea, J.A. Toca-Ramírez, C. Lemus, J. Toca-Ramírez, and M. Alonso-Spilsbury, “Horse Meat for Human Consumption in México: Slaughter Performance and Carcass Morphometry”, *Journal of Food Technology*, 106-110.

slaughter will probably have ingested, or been treated or injected with, multiple chemical substances that are known to be dangerous to humans, are untested on humans, or are specifically prohibited for use in animals raised for human consumption.

The Mexican government regulates the presence of toxic residues in the meat of animals for human consumption by the enactment of the Federal Law for Animal Health (*Ley Federal de Sanidad Animal de México*). In 2014, to comply with international toxic residue requirements, the Mexican Ministry of Agriculture (Sagarpa) adopted an agreement establishing maximum levels of toxic and contaminating residues.¹⁰ The FAO also issues guidelines on maximum residue limits (MRL) on cattle.¹¹

However, horses are gathered from random sources at various stages in their lives. There is no effective system either in the U.S. or in Mexico to track medications and veterinary treatments given to horses to ensure that their meat is safe for human consumption.¹² This paper aims at moving forward the research agenda on horsemeat consumption and meat mislabeling practices in Mexico, through a collection of case studies from various Mexican cities. More research will be needed in the future to attain a comprehensive assessment of this phenomenon in Mexico.

II. OBJECTIVES

This study has three main objectives. The first is to understand the dynamics of selling horse meat in Mexico through surveys in various outlets where meat is sold in cities across the country where the breeding, slaughter, sale and consumption of this type of meat are known.

¹⁰ Secretaría de Gobernación, *Diario Oficial de la Federación*, “Acuerdo por el que se establecen los criterios para determinar los límites máximos de residuos tóxicos y contaminantes, de funcionamiento de métodos analíticos, el Programa Nacional de Control y Monitoreo de Residuos Tóxicos en los bienes de origen animal, recursos acuícolas y pesqueros, y Programa de Monitoreo de Residuos Tóxicos en animales, así como el módulo de consulta, los cuales se encuentran regulados por la Secretaría de Agricultura, Ganadería, Desarrollo Rural, Pesca y Alimentación”, 9 October, 2014.

¹¹ Food and Agriculture Organization of the United Nations, *Codex Alimentarius Commission. Maximum Residue Limits for Veterinary Drugs in Foods (Updated as at the 35th Session of the Codex Alimentarius Commission)*, Rome, July 2012. ftp://ftp.fao.org/codex/weblinks/MRL2_e_2012.pdf

¹² European Commission, *Final report of an audit carried out in Mexico from 24 June to 04 July 2014 in order to evaluate the operation of controls over the production of fresh horse meat and meat products intended for export to the European Union, including monitoring of residues and contaminants as well as certification procedures, DG(SANCO), 2014-7223 – MR FINAL*, 2014.

The second objective is to detect the fraudulent sale of horse meat as beef through meat sampling (in the same outlets that were interviewed) with subsequent DNA analysis. The third one is to detect potential toxic residues (specifically by a clenbuterol test) in the samples that tested positive for equine flesh.

III. METHODOLOGY

1. Cities selected for the Study

Six Mexican cities were selected for the study: Aguascalientes, Chihuahua, and Zacatecas, where horse slaughter is conducted in TIF (Federally Inspected Slaughterhouses)¹³; San Vicente Chicoloapan (SVC), where the slaughter of horses takes place in municipal plants; Mexico City, the most populated city in the country; and Pachuca, a city that was chosen randomly.

In order to determine within each city the places of sampling and putting in place of questionnaires, each one of the five cities was divided into four quadrants, which were classified as Zone A (West-North), Zone B (East-North), Zone C (West-South) and Zone D (East-South). In cities with irregular dispersion and concentration of businesses, quadrants were reported only as Zone North (North-West and North-East) and Zone South (South-West and South-East).

2. Establishments for the sampling

In each of these zones (A-D), five types of establishment were selected: 1) Fixed and established markets; 2) Mobile street markets or semi-fixed markets on wheels, not established; 3) Supermarkets or department stores that retail meat; 4) Fixed and established butcher shops; and 5) Mexican *tapas* selling stands fixed or semi-fixed, plus established snack bars.

¹³ It is worth noting that two of the selected cities (Aguascalientes and Zacatecas) are located within the Western region of Mexico, where the production of horse meat is the largest of the country. Financiera Rural, *Carne de caballo. Contexto nacional*, September 2012. The report is no longer available at the website of the institution: <https://www.gob.mx/fnd>.

3. Types of samples

The types of samples collected in each of the establishments included various products depending on the selling site. The research team purchased ground beef and Milanese steaks in supermarkets and butcher shops. In addition, the team obtained the following meat samples in markets, street markets, and snack bars: *tacos* with grilled or fried steaks, fried *tacos* with shredded cooked meat, stewed *gorditas*, and *burritos*, the latter two mainly in the northern cities of the country. Photographs of the samples are visible in the Photographic Annex 1.

4. Size of samples

The sample for raw meat was 200 g each. In the case of the cooked samples, three to six units were purchased to obtain a sample of 50-100 g each.

5. Collection and shipment of samples

Once the research team mapped and allocated zones for each city, they defined the number of businesses to sample per zone to reach a total of 100 samples. This number had to be adjusted in each city mainly due to the insufficient or excessive offer of these products at each site. Then, the team collected samples at selected sites and at the same time, put in place surveys in all businesses present in each area.

During the sampling period, meat samples were stored in insulated containers with cooling at a temperature below 8°C. The samples were then packed in plastic bags, identified, and sealed. The bags remained within an isothermal container at a temperature of 4°C. Once all samples were collected for the city, these were shipped sealed and identified to the Laboratory of Meat Quality of the ITSON.

6. Establishments where meat was purchased for DNA analysis

In the Annex 1, all places where sampling was carried are listed per zone. In Table 1 there is a summary of the surveyed places per city and of the samples collected for DNA analysis. In total, there were 157 establishments in six cities.

Table 1. Visited outlets where surveys were made and/or where samples were collected.

	Mexico City	Zacatecas	Aguascalientes	Chihuahua	Pachuca	SVC	TOTAL
Markets	10	9	14	2	11	2	48
Supermarkets	9	4	10	4	0	2	29
Street markets	11	7	7	4	0	4	33
Butcher shops	10	12	5	6	4	2	39
Others	0	0	3	0	5	0	8
TOTAL	40	32	39	16	20	10	157

7. Interviews carried

Researchers in this study made two types of interviews:

- 1) Specific in places where there was horsemeat in sight for sale. In this case, researchers noted information regarding the zone, the outlet’s address and type and the survey taker. Data was also collected concerning the type of meat, if it was labeled, the price, etc. The model of this survey is presented below:

Date
Zone: NW/NE/SW/SE
Retail outlet:
Address:
Interviewer:
HORSEMEAT ON SIGHT

Is it clearly labelled?
Which cuts are sold?
What is the price per kilogram and cut type?
Where does the horsemeat come from?
Who is your supplier?
Is it highly valued by the general customers?
Which kind of customer asks for horsemeat?
How it is usually cooked?
QUICK ASSESSMENT OF THE PLACE ON SANITARY CONDITIONS
Any other comments on the place

Fecha:
Zona: NO/NE/SO/SE
Tipo de Punto de venta:
Dirección:
Encuestador:

SI HAY CARNE DE CABALLO A LA VISTA

¿Esta etiquetada como tal?
¿Qué tipo de cortes se vende?
¿Cuál es el precio/ kg / tipo de corte?
¿De dónde es la carne de caballo?
¿Quién es su proveedor?
¿Es muy apreciada por el público?
¿Qué tipo de consumidor requiere dicha carne?
¿Cómo la cocinan normalmente?

HACER UNA EVALUACION RAPIDA DEL LUGAR EN CUESTION SANITARIA

Cualquier otra anotación sobre el lugar:

- 2) Researchers conducted non-specific interviews in places where horsemeat was not displayed, or where the sale of horse meat was not advertised. In these places, information was taken regarding the zone, the outlet's type, address, and the survey taker. Respondents were also asked why no horsemeat is being offered, if there are known providers, etc. The model of this survey can be found below:

Date
Zone: NW/NE/SW/SE
Retail outlet:

Address:
Interviewer:
NO VISIBLE SIGN OF HORSE MEAT
Why do you not offer horsemeat?
Do you know any supplier to acquire it?
Would you like to sell horsemeat? Why?
QUICK ASSESSMENT OF THE PLACE ON SANITARY CONDITIONS
Any other comments on the place

Fecha:
Zona: NO/NE/SO/SE
Tipo de Punto de venta:
Dirección:
Encuestador:

NO HAY CARNE DE CABALLO A LA VISTA
¿Por qué no ofrece carne de caballo?
¿Conoce a algún proveedor para adquirirla?
¿Usted quisiera vender carne de caballo? Porque?
HACER UNA EVALUACION RAPIDA DEL LUGAR EN CUESTION SANITARIA
Cualquier otra anotación sobre el lugar:

8. DNA extraction and PCR test for determining the species of origin

This methodology was used as reported by Hernández *et al.*, with some modifications for processed products.¹⁴ The reagents used in the extractions: Tris-HCl (hydroxymethylaminomethane), sodium dodecyl sulfate (SDS), ethylenediaminetetraacetic acid (EDTA), Chloroform-Isoamyl alcohol (24:1), N-acetyl-N, N, N-trimethyl ammonium chloride (CTAB) balanced Phenol, ethanol, 2-hydroxyethyl cellulose (HEC) PM 90,000 and poly (vinyl alcohol) (PVA) PM 50,000 were from SIGMA (U.S.); Proteisana K and RNase A were manufactured by Invitrogen (Mexico).

¹⁴ J.F. Hernández, A.F. González-Córdova, R. Rodríguez-Ramírez y B. Vallejo-Córdoba, “Development of a polymerase chain reaction and capillary gel electrophoresis method for the detection of chicken or turkey meat in heat-treated pork meat mixtures”, *Analytica Chimica Acta*, 708 (2011), 146-154.

Researchers performed an initial incubation of the processed and raw samples (100 mg) in 500 µl of buffer homogenization (0.4 M NaCl, 10 mM Tris-HCl, 2 mM EDTA, pH 8.0) for 15 min at 55 ° C. Homogenization was carried out in a Turrax homogenizer at 11.0000 50 rpm for 45 seconds. Ten ml RNase (10 mg/ml) was added to the homogenate and incubated for one hour at 37 ° C. Subsequently, 10 ml of proteinase K (20 mg/ml) and 50 ml of SDS at 20% were added, incubating again for one hour at 60° C. The mixture was centrifuged for 10 minutes at 12,000 x g at 4° C. To the supernate were added 300ml 6M NaCl and 88 ml of CTAB at 10% and incubated at 55° C for one hour. Later, a volume of 850 ml of phenol:chloroform:isoamyl alcohol 25:24:1 was added. Subsequently, the aqueous phase was recovered, where the DNA molecules can be found and an equal volume of isopropanol was added and incubated at least one hour at -20° C. Then it was centrifuged for 40 minutes at 12,000 x g at 4° C. The DNA remained in the precipitate, which was washed twice with 300 ml of ethanol at 70% to fix it (centrifuged for 2 minutes at 12,000 x g at 4° C each). Technicians then proceeded to evaporate the ethanol for 10 minutes. Once evaporated, 100 ml of TE buffer (10 mM Tris-HCl, 1 mM EDTA, pH 8.0) were added to the sediment.

Prior to DNA amplification by PCR, the DNA extracted was found to be sufficiently pure and free of protein contamination. Technicians used a spectrophotometer Cary 100™ (Agilent) under the following indicators to measure the concentration of DNA: the DNA presents a maximum absorbance at 260 nm (50 µg/mL, have an OD at 260=1), while proteins have it at 280 nm. The purity was calculated, taking it into account between A260/A280. For results from 1.9 to 1.7, the DNA is considered pure.

Lab technicians performed the amplification of specific fragments by standard PCR and QC-PCR. The mtDNA amplification by PCR was carried out considering a final volume of 50 mL. For the amplification of the selected sequences, a thermocycling program Eppendorf™ (Malaysia) was used, which consisted of one initial denaturation step in which the reaction mixture was maintained at 94° C for 3 minutes so that the two chains of DNA mod would separate. Subsequently, the reaction mixtures were subjected to 32 cycles of three steps each (denaturation, alignment and extension) as shown below.

Reaction step	Temperature	Time	Number of cycles
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Denaturalization	94 °C	3 min	1
Denaturalization	94 °C	1 min	32
Alignment	56 °C	45 sec	
Extension	72 °C	45 sec	
Final extension	72 °C	10 min	1
Refrigeration	4 °C	Stand by	1

Once the reactions finished (2.40 min), 10-15 µl of the PCR products of the samples were taken to be analyzed by conventional electrophoresis. The nucleotide sequences of the horse species used in this research were designed using the program PRIMER3 (<http://bioinfo.ut.ee/primer3/>). The specific sequence for horses obtained from GenBank (<http://www.ncbi.nlm.nih.gov/genbank/>) was the mitochondrial Cit-b gene.

The similarity of the selected sequences was analyzed using the computer program BLAST® (<http://www.ncbi.nlm.nih.gov/BLAST>). The selected sequences for the design of oligonucleotides were synthesized in the laboratory ProbioTek (Monterrey, Mexico) as shown below:

Species	Primer	Sequence 5'-3'	Gene
Horse	PFw1-Eqca	CTACATCGGTA CTACCCTCGTC 22 pb	Mitochondrial Cit-b
	PRv-Eqca	AATGTACGACT ACCAGGGCTG 21 pb	

9. Statistical analysis

The results obtained were subjected to a simple statistical frequency and percentages in SPSS (2010).

IV. RESULTS

IV. 1. Results of surveys on knowledge of selling horse meat

Table 2 presents the sampled establishments in each city and the visited outlets of horsemeat, the total number of acquired samples and samples that we were told were horsemeat.

Table 2. Surveys conducted per city.

	Non-specific surveys	Specific surveys (Horse meat shops)
Mexico City	108	0
Aguascalientes	77	1*
Zacatecas	40	1
Chihuahua	76	3
Pachuca	20	0
San Vicente Chicoloapan	10	0
Total	334	5

**No horse meat was offered as such, it was during the survey that the offer was made.*

Researchers with this study carried out the sampling randomly in the cities. However, researchers also visited establishments that officially sold horsemeat (Zacatecas, Chihuahua, and Aguascalientes) and some that are also unofficially known (only in Mexico City) as places selling horse meat. Overall, 339 surveys were carried out. The overwhelming majority (334) were not specific because they were places that were not selling horsemeat. Only five surveys were specific because they were places where the product is officially sold. The results of both surveys per city are below.

1) Mexico City

In Mexico City, the research team found no horsemeat outlets, therefore 108 non-specific surveys were conducted to business owners across the city. The results thereof are shown in Figures 1-4.

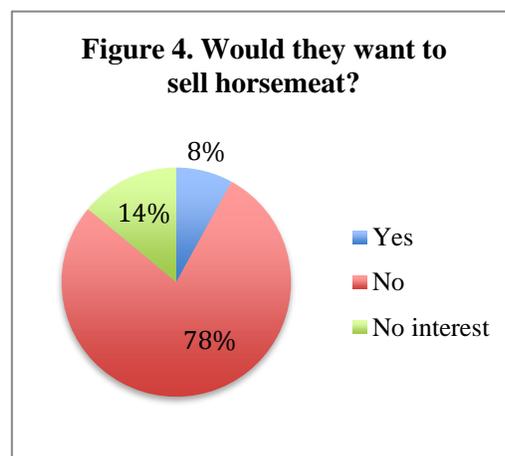
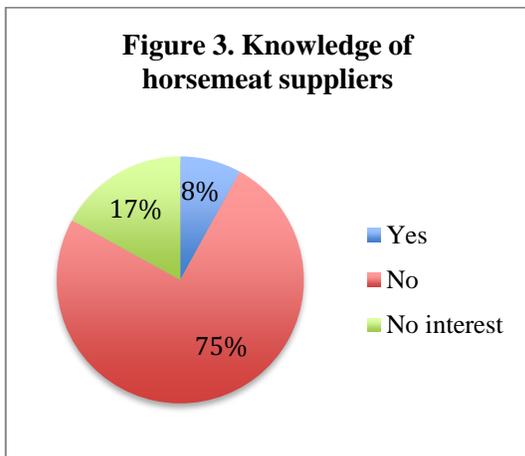
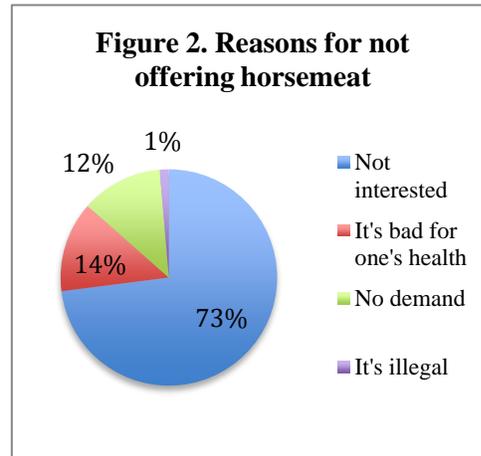
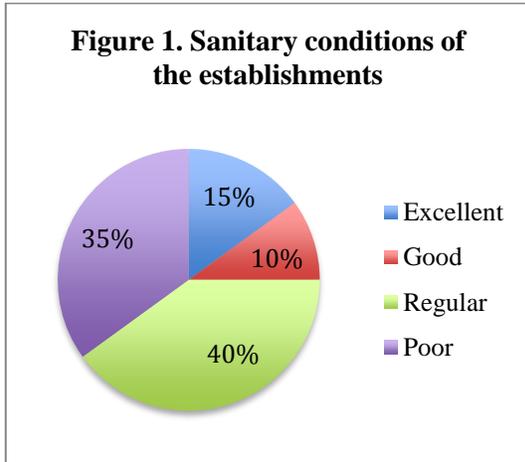


Figure 1 shows that in Mexico City, 15% of establishments are in excellent conditions and these represent mostly sampled supermarkets. On the other hand, half the establishments, such as butcher places and markets, have sanitary conditions categorised between good and regular. The remaining 35% of the surveyed establishments, normally the street vendors and some markets, have poor sanitary conditions. Figure 2 shows the reasons why horsemeat was not sold and reveal that around two-thirds of respondents were not interested, while the rest considered it bad for the health, illegal, or because there is no demand. Figure 3 shows that 75% of people did not know any suppliers of horsemeat while Figure 4 shows that almost 80% were not even interested in selling it.

2) Zacatecas

In the city of Zacatecas, researchers found only one outlet openly selling horse meat (for which a specific survey was done), making a total of 40 surveys to business owners throughout the city (non-specific surveys). These results are shown in Figures 5-8.

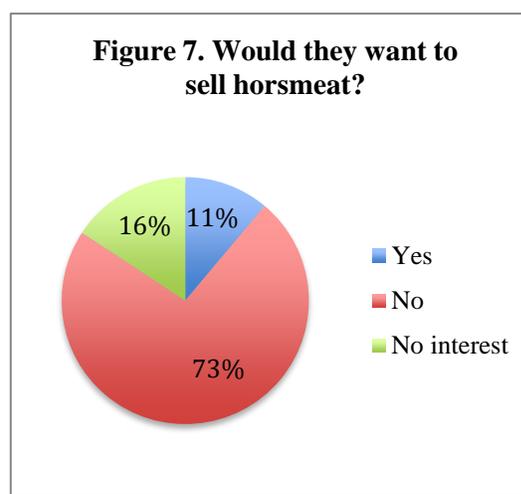
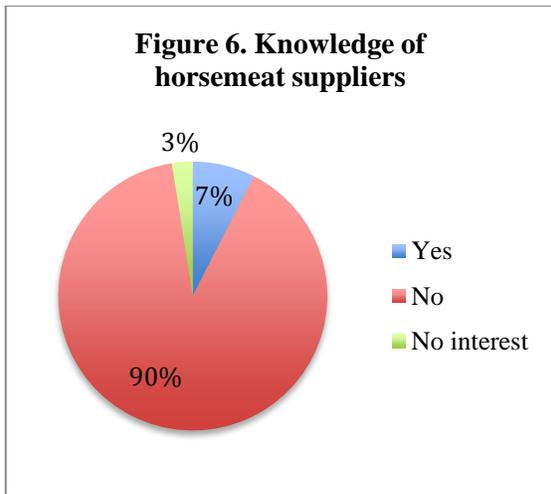
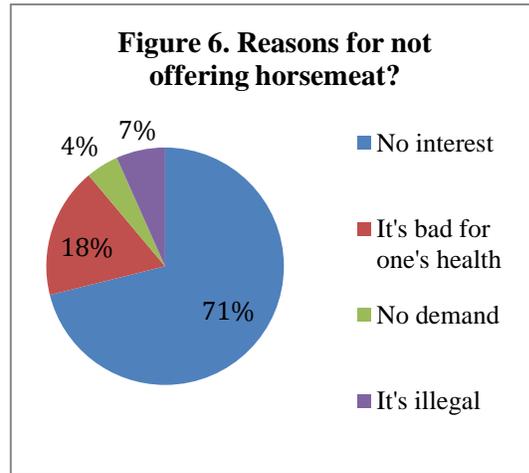
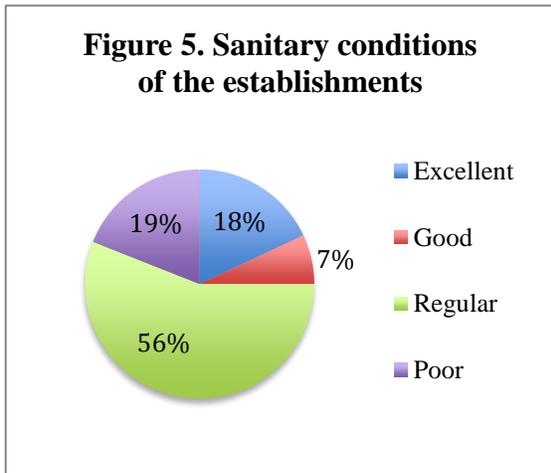


Figure 4 shows that health conditions were excellent in supermarkets and butcher shops, regular in markets and good and bad in street markets. In figures 5, 6, and 7, results were similar to those obtained in Mexico City showing a huge lack of interest in commercializing this kind of meat in their establishments.

In the Zone North (North-West) of the city of Zacatecas, there is one distributor of previously vacuum-packed and labelled horsemeat, “Cavalli”. From this place, the research team did not collect any samples for a DNA test. First, because the meat was openly offered as horsemeat and because they do not sell quantities required (as sample size is defined in the methodology). Products offered for sale were fresh meat such as steak, solid piece, special ground (90-10), commercial ground (80-20), and marinated and sewed steak (*carne enchilada*), as well as frozen as pocketed T-bone chop (only non vacuum-packaged product) and burger-ready medallions for roasting or deep-frying, tripes, and livers for taco bars. Jerky meat as a snack and canned meat for pet food were also offered for sale.

All products come from the TIF plant in the town of Fresnillo (state of Zacatecas) where an average of 1,800 horses are killed per month. The distributor is one of the owners of the packing plant that processes in the slaughterhouse. The distributor commented that their sales were limited due to the EU suspension, which explains why two outlets were set up in the city of Zacatecas, one in the Condesa area managed by the owners of the plant and another in the centre of Zacatecas (the site was not found active) which distribute the product directly to the final consumer.

In 2015, a panel of specialists involved in the production, slaughtering, and distribution of meat of these species discussed on the local media (radio and television) the advantages and disadvantages of horsemeat consumption. Their favourable view towards equine meat spread through the city. As a result, sales increased by 30% from the previous year. The activity of the *Empacadora de Carnes de Fresnillo, SA de CV* is focused on the collection of equines from across the state of Zacatecas and its surroundings. As for Durango, Coahuila, and San Luis Potosí, these horses represent only 10% of the total of the animals slaughtered, the remaining 90% are horses imported from the U.S. for meat export to countries such as Vietnam, Japan, and Russia, mainly in addition to sales via outlets in the cities of Fresnillo and Zacatecas. The whole of the animal is used since in addition to the meat obtained, other by-products are made such as pet food, dried meat, hides, and hairs for the confection of brushes, paintbrushes, and leather goods.

The main outlets' buyers are homemakers, because according to the distributor, vendors of convenience food and snacks at street markets purchase their meat mainly from

suppliers that commercialise horse carcasses obtained in municipal or clandestine slaughterhouses.

3) Aguascalientes

In the city of Aguascalientes, researchers with this study found only one outlet selling horse meat (for which a specific survey was done), and visited 77 meat establishments where non-specific surveys were conducted with business owners throughout the city. These results are shown in Figures 9-12.

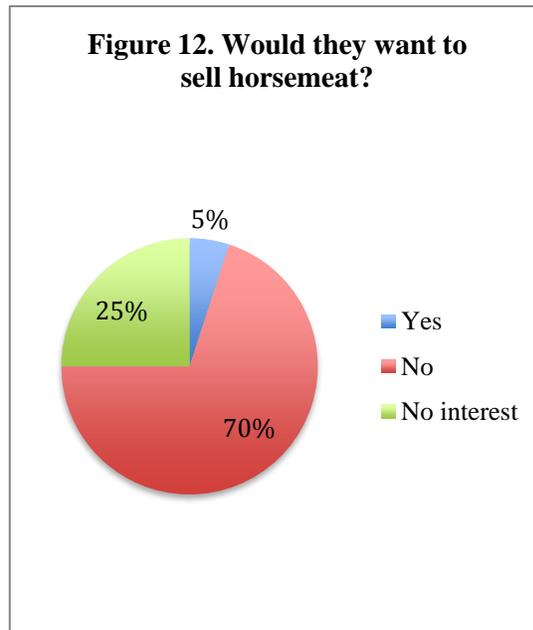
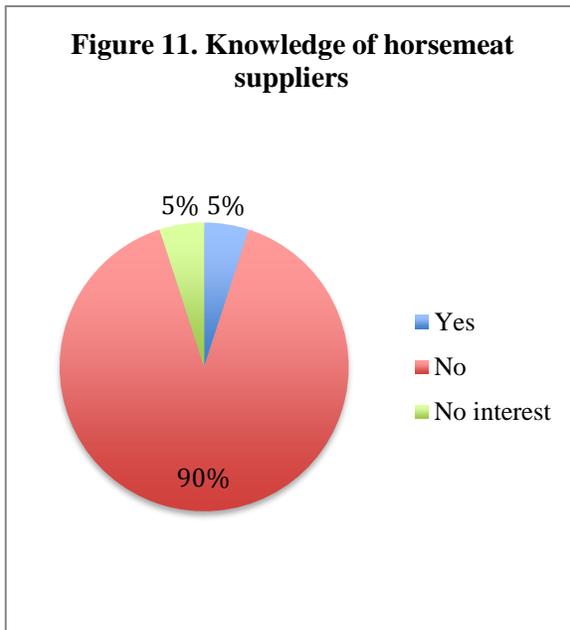
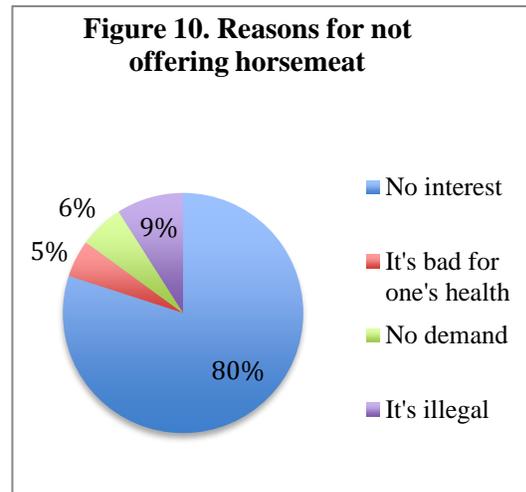
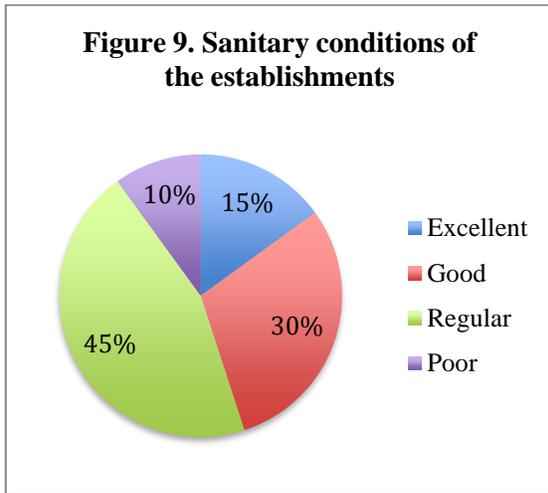
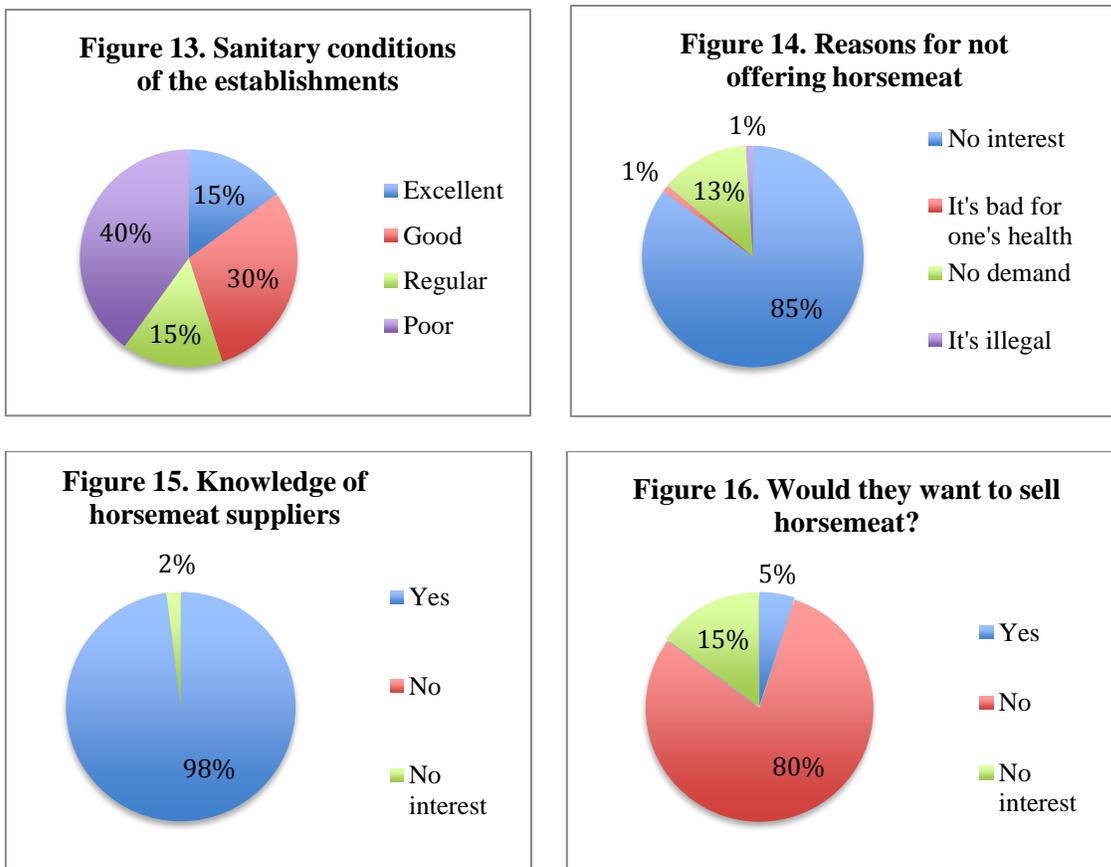


Figure 9 shows the establishments of this city overall have a better hygiene, with the exception of the street markets, which are categorised under poor hygiene conditions. In general, in Aguascalientes, meat distributors are not interested in selling horsemeat (Figures 10, 11 and 12).

4) Chihuahua

In the city of Chihuahua, researchers in this study sampled 79 establishments, with only three shops openly selling horsemeat. In the other 76 establishments visited, non-specific surveys were applied to business owners throughout the city. These results are shown in Figures 13-16.



In Figure 13, supermarkets score excellent conditions while for other establishments conditions are categorised from good to bad mainly because butchery products are mixed with other products, such as groceries and greengroceries. The lack of interest in selling horsemeat is high (Figures 14 and 16). However, it is worth noting that most outlets know horsemeat suppliers, which makes Chihuahua a special city in this regard (Figure 15). The

butcher shops that sell horsemeat from a TIF plant are found in the 4th Street in the center of the city of Chihuahua. In a radius of not more than 100 meters, four other butcher shops also sell this type of meat passing it for cheap beef, according to the manager of the municipal slaughterhouse Sanher.

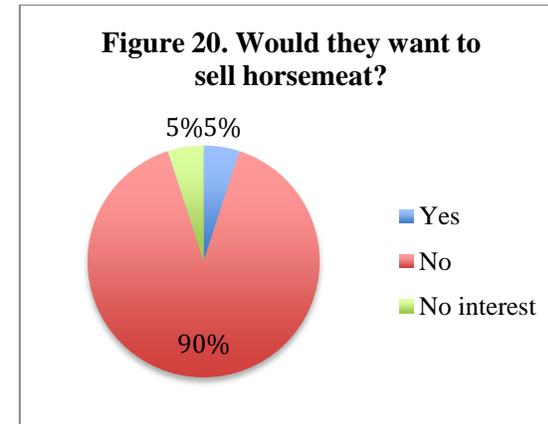
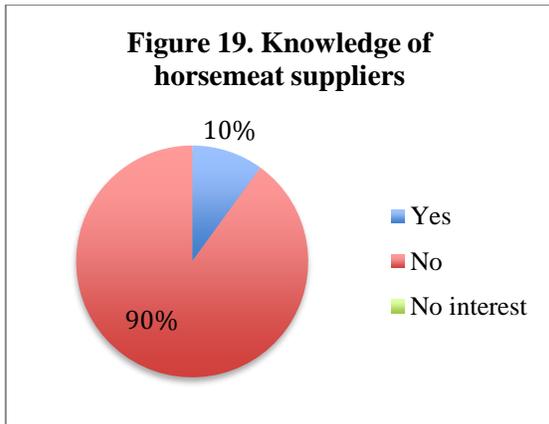
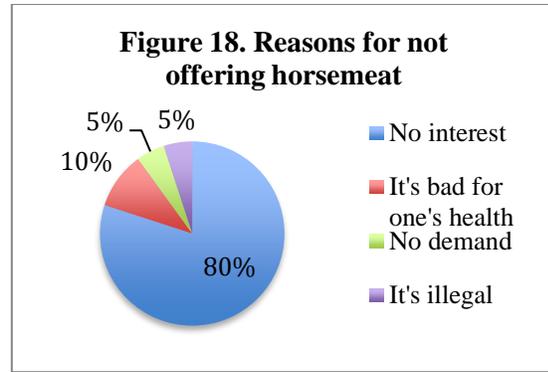
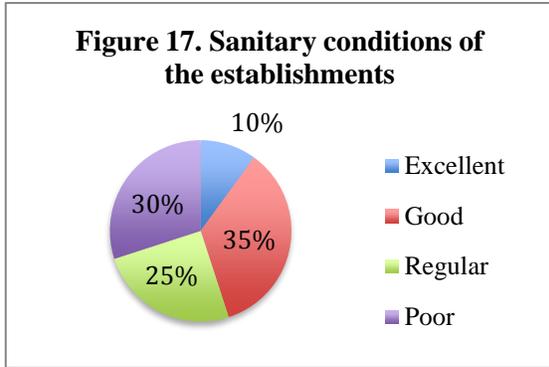
In contrast to the results from other cities where sampling was carried out, the sale of equine meat in Chihuahua is widespread and accepted. Data were even collected for the two visited slaughterhouses.

In the TIF-366 plant, authorized for the slaughter of cattle, pigs and horses, located in the eastern part of the city, on average 2,200 horses are killed per month. This figure can increase by 40% in the winter and 20% at the beginning of the dry season in order to prevent animals from losing weight due to the lack of food. According to the slaughterplant manager, a large majority of the horses slaughtered in this plant originate from the U.S. The meat from this plant is exported to mainly Asian countries, with only a small portion distributed in Juarez City. A few of the horses killed are from the Chihuahua mountains or from neighbouring states (mainly Durango). The meat of Mexican origin is distributed throughout the city in various establishments that sell equine meat. Vendors sell such meat for local consumption, either raw or prepared in multiple ways, without mentioning its equine origin.

Also, the research team visited the municipal slaughterhouse Sanher that is authorized to slaughter horses and cattle on the Juan Pablo Segundo road, in the Eastern part of the city. According to the manager, an average of 350 horses are killed per month with an increase of 15% in the dry season and 25% in the winter. Horses originate from the mountains of the same state and to a lesser extent, from Durango. The meat is exclusively destined to supply the city's businesses, which sell both raw meat and cooked in various forms.

5) *Pachuca*

In the city of Pachuca, the research team sampled 20 establishments and found no shops selling horsemeat. Therefore, 20 non-specific surveys were conducted with business owners in the city. These results are shown in Figures 17-20.



In Pachuca, the sanitary conditions of 55% of the establishments surveyed are categorised from regular to bad (Figure 17). Figures 18, 19 and 20 reflect the lack of knowledge on this subject and a lack of interest from shop managers and owners in selling this type of meat.

6) San Vicente Chicoloapan

In the city of San Vicente Chicoloapan, the research team sampled 10 establishments and found no shops selling horsemeat. Therefore, 10 non-specific surveys were conducted with business owners of the city. These results are shown in Figures 21-24.

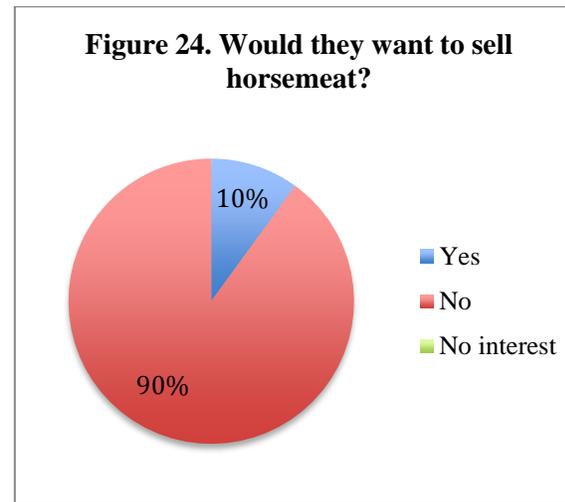
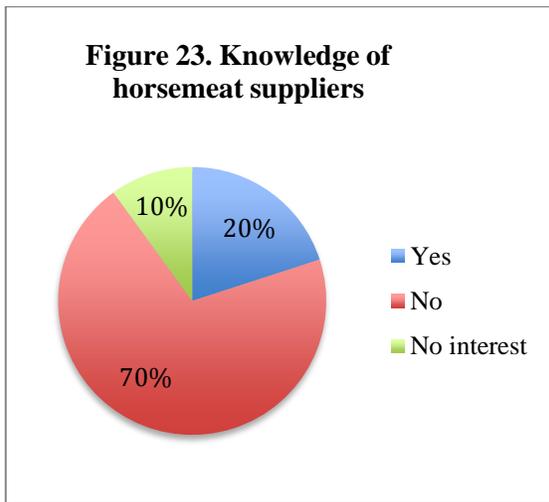
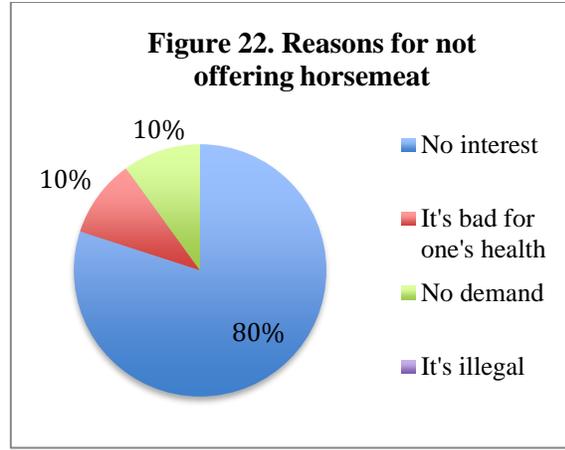
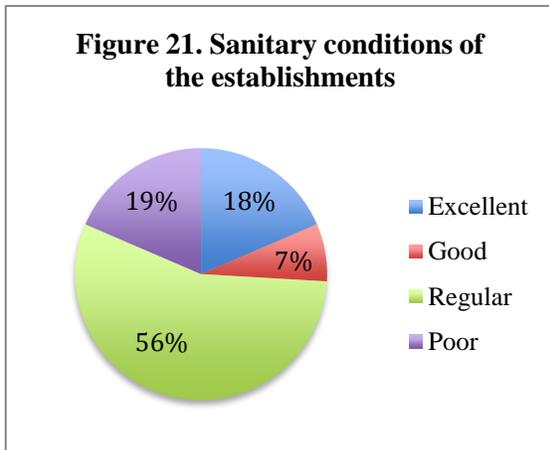


Figure 21 shows that 75% of establishments surveyed have regular to poor hygiene. During the sampling, researchers found that established shops have a better hygiene, while street markets had the worst ratings in this category. As in most of the cities surveyed, there is also a high lack of interest from business owners in offering and distributing horsemeat in their establishments (Figures 22, 23, and 24).

IV. 2. Results of PCR in DNA samples of raw and cooked meat

Through the molecular PCR technique, out of the 433 samples collected in different cities mentioned in the methodology, 43 tested positive for horse DNA, accounting for 9.93% (Table 3). Below are the results of the distribution of positive samples per city, depending on the type of sample and establishment where it was purchased.

Table 3. Samples selected and analyzed per city.

	Total samples collected	Samples collected that are horse meat
Mexico City	96	(0)
Aguascalientes	74	(4)
Zacatecas	56	(0)
Chihuahua	133	(5)
Pachuca	34	(0)
SVC	40	(0)
Total	433	(9)

* No horse meat was offering as such, it was during the survey that the offer was changed.

Table 4. Samples testing positive to horse DNA per city.

	Total samples collected // Samples collected in establishments openly selling horse meat –in parenthesis ().	Samples positive to horse DNA	Percentage of positive samples
Mexico City	96 (0)	12	12.5
Zacatecas	56 (0)	4	7.14
Aguascalientes	74 (4)	8	10.81
Chihuahua	133 (5)	15	11.28
Pachuca	34 (0)	4	11.76
SVC	40 (0)	0	0
Total	433 (9)	43	9.93

Table 4 reveals that the highest percentages of positive samples (between 10 and 13%) were found in four of the surveyed cities (Aguascalientes, Chihuahua, Mexico City, and Pachuca). A lower percentage was reported in Zacatecas and none at all in San Vicente Chicoloapan.

Table 5. Distribution of samples positive for horse meat by type of meat.

	Samples analyzed	Positive samples (horse meat)	Percentage of positive samples
Steak	146	10	6.85
Ground	150	18	12.00
<i>Tacos</i>	62	8	12.90
<i>Burritos</i>	14	4	28.60
Others	61	3	4.92
TOTAL	433	43	9.93

Table 5 shows that the *burritos* were in proportion the type of samples where other meats were most commonly mixed with horsemeat (28.60%).

Table 6. Distribution of samples positive for horsemeat by type of establishment.

	Samples analyzed	Positive samples (horsemeat)	Percentage of positive samples
Supermarket	70	0	0
Market	106	11	10.38
Butcheries	146	17	11.64
Street vendor	78	9	11.54
Other	33	6	18.20
TOTAL	433	43	9.93

Table 6 shows no samples positive for horsemeat were found in the supermarkets surveyed, while the distribution of positive samples tends to be similar for other establishments. The

total number of samples that tested positive for horse DNA in each city according to the type of establishment and type of sample is as shown next.

IV.3. Positive results by city, type of meat and establishment

1) Mexico City

In Mexico City, the research team analyzed 96 samples, 12 of which tested positive for horsemeat, i.e. 12.5% (Table 7). The only zone in Mexico City where no positive samples were found was the North-West. In the North-East one positive sample was found, while two and seven positive samples were respectively collected in the South-West and the South-East zones.

Table 7. Distribution of samples positive for horsemeat in Mexico City

Distribution of samples positive for horsemeat in Mexico City					
Type of meat	Number	%	Purchase Location	Number	%
Steak	2	17	Supermarket	0	0
Ground	5	42	Market	4	33.3
Tacos	4	33	Butcher shop	3	25
Crispy <i>tacos</i> / <i>burritos</i>	1	8	Street market	4	33.3
Other	0	0	Snack and <i>tapas</i> bar	1	8.4

2) Aguascalientes

From Aguascalientes, researchers analyzed 74 samples and eight of them were positive for horsemeat, that is to say 10.81% (Table 8).

Table 8. Distribution of samples positive for horsemeat in Aguascalientes.

Distribution of samples positive for horsemeat in Aguascalientes					
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Type of Meat	Number	%	Purchase Location	Number	%
Steak	1	12.5	Supermarket	0	0
Ground	5	62.5	Market	5	62.5
<i>Tacos</i>	2	27	Butcher shop	1	12.5
Crispy <i>tacos/burritos</i>	0	0	Street market	2	25
Other	0	0	Snack and <i>tapas</i> bar	0	0

3) *Zacatecas*

In Zacatecas, the team collected 56 samples, with four of them testing positive for horsemeat, i.e. 7.14% (Table 9).

Table 9. Distribution of samples positive for horsemeat in Zacatecas.

Distribution of samples positive for horsemeat in Zacatecas					
Type of Meat	Number	%	Purchase Location	Number	%
Steak	0	0	Supermarket	0	0
Ground	3	75	Market	1	25
<i>Tacos</i>	0	0	Butcher shop	3	75
Crispy <i>tacos/burritos</i>	1	25	Street market	0	0
Other	0	0	Snack and <i>tapas</i> bar	0	0

4) *Chihuahua*

In Chihuahua, out of 133 samples collected, 15 appeared to be horsemeat, that is, 11.28% (Table 10).

Table 10. Distribution of samples positive for horsemeat in Chihuahua

Distribution of samples positive for horsemeat in Chihuahua					
Type of Meat	Number	%	Purchase Location	Number	%
Steak	6	40	Supermarket	0	0
Ground	5	33.3	Market	0	0
<i>Tacos</i>	1	6.7	Butcher shop	10	73.3
Crispy <i>tacos/burritos</i>	2	13.3	Street market	3	20
Other	1	6.7	Snack and <i>tapas</i> bar	1	6.7

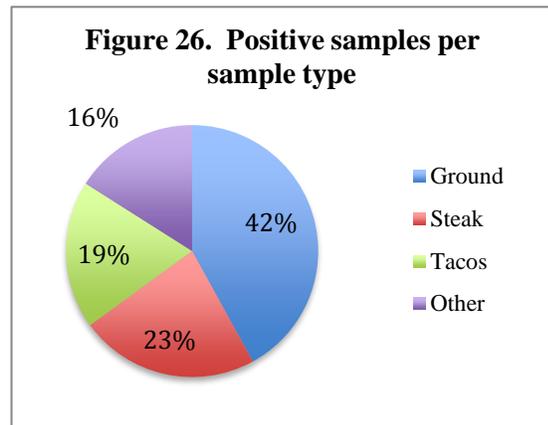
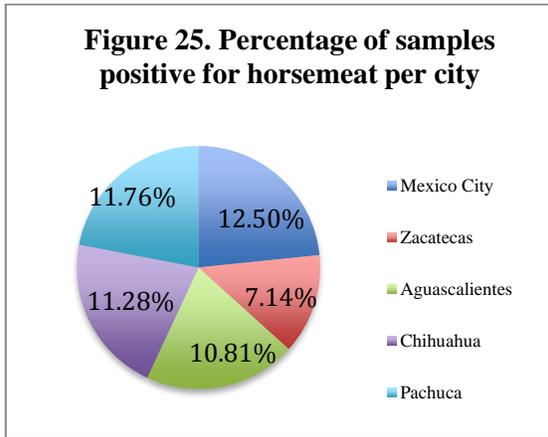
5) *Pachuca and San Vicente Chicoloapan*

In Pachuca, out of 34 samples collected, four tested positive for horsemeat, that is to say 11.76% (Table 11). In San Vicente, all 40 samples collected tested negative.

Table 11. Distribution of samples positive for horsemeat in Pachuca

Distribution of samples positive for horsemeat in Pachuca					
Type of Meat	Number	%	Purchase Location	Number	%
Steak	1	25	Supermarket	0	0
Ground	0	0	Market	1	25
<i>Tacos</i>	1	25	Butcher shop	0	0
Crispy <i>tacos</i>	0	0	Street market	0	0
Other	2	50	Snack and <i>tapas</i> bar	3	75

In short, and as shown in Figure 25, out of the total positive samples per city, 12.5% in Mexico City, 11.76% in Pachuca and similar numbers were found in Chihuahua with 11.28%, Aguascalientes 10.81% and 7.14% in Zacatecas.¹⁵



Out of all the samples that tested positive for horsemeat, 42% appeared to be ground meat, 23% were steaks, while *tacos* and shredded meat used in *burritos* and other food items represented respectively 19% and 16% of positive samples. It is relevant to consider that 77% of the positive samples were types of processed meat (ground meat, *tacos*, *burritos*, and others).

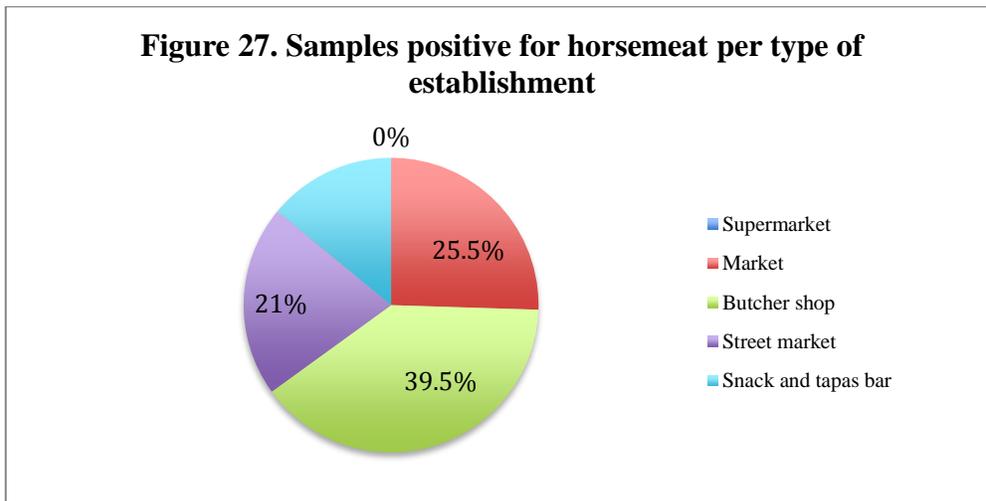


Figure 27 shows that, apart from supermarkets, samples that tested positive for horse DNA were found in all types of establishments surveyed. The use of horsemeat as a substitute for

¹⁵ The number of samples was selected accordingly to the population and consumption rates.

beef or other meats appears therefore to be widespread in markets, butcher shops, and street markets of the cities where samples were collected.

IV.4. Clenbuterol analysis

As for the samples that tested positive for horsemeat (43), a further study was carried out at Noxgen Laboratory in Mexico to identify a specific potential toxic residue (clenbuterol) in the aforementioned flesh. The available devices at the laboratory were of no use on cooked meat and thus they permitted testing only on the raw meat samples (29) that originated in four of the original six cities (Aguascalientes, Zacatecas, Chihuahua, and Mexico City).

Clenbuterol or clenbuterol hydrochloride is a β -agonist belonging to the adrenergic group. Its chemical name is 1-(4-amino-3,5-dichlorophenyl)-2-(tert-butylamino) ethanol and it has a chemical structure similar to catecholamines like adrenaline and noradrenaline. Initially, it was used in veterinary medicine due to its bronchodilator, decongestive, and tocolytic properties, but it was later discovered that a dose ten times the therapeutic one had an anabolic effect, speeding up the synthesis of protein and reducing that of fat, and is the reason why this group has been dubbed “energy distribution agents”.

Clenbuterol is absorbed by nearly all the tissues due to the location of the β -adrenoceptors. Most research into clenbuterol residues in cattle have found that these tend to be concentrated in the eyes, liver, kidneys, and lungs, while the lowest levels are found in muscle and fat.¹⁶ The detection method only detects the drug in its pure form, since its metabolites are not of toxicological importance.

The use of clenbuterol in food animals has potential adverse human health effects. The Food Animal Residue Avoidance Databank (FARAD) has placed restrictions on its use. Currently, clenbuterol has been used in food animals “to increase weight gain and lean body mass. High doses of the drug are necessary for these effects, which may have been one of the factors that led to the reported hospitalization of more than 1,200 people and the death of three people in France and Spain that were linked to clenbuterol residues in the liver of illegally treated animals”. Furthermore, numerous humans have developed symptoms of

¹⁶ L. Sumano, H., L. Ocampo C. and L. Gutiérrez O., “Clenbuterol and Other β - Agonists. An Option for Cattle Production or a Health Risk?”, *Revista Veterinaria México* 33 (2002), 137-159.

toxicosis, with muscle tremor, tachycardia, and heart palpitations being the most commonly reported.¹⁷

According to FAO, the maximum daily admissible value of clenbuterol is 0.240 micrograms for a 60 kg person. In slaughter animals, the maximum residue limit (MRL) for the drug, both in cattle and horses, is 0.2 micrograms/kg for muscle and fat, 0.6 micrograms/kg for the liver and kidneys, and 0.05 micrograms/liter for cattle milk.¹⁸ Liver is the edible tissue that contains the highest residue concentrations and cooking only decreases drug residues minimally.¹⁹

As a result, some countries have banned the use of clenbuterol in cattle for human consumption. In Mexico, there are three general regulations that explicitly ban clenbuterol for animals destined for human consumption:

1. *NOM-194-SSA1-2004 Health Specifications for Establishments that Slaughter and Skin Animals for Food, Storage, Transportation and Sale*, which states that “cattle products should be free of clenbuterol” and that “the enzyme-linked immunosorbent assay method” should be used to detect it (which was the method used for this study).
2. *NOM-061-ZOO-1999, Health Specifications for Animal Feed*, which stipulates that “the use of clenbuterol is banned, along with all ingredients and/or food additives that have been proven to pose a potential threat to public health or pose a health risk and for which there is no technical evidence to support their use in animal nutrition.”
3. *NOM-065-ZOO-2003, Technical Specifications for the Eradication of the Use of Non-Authorized Beta-Agonists in Animals*, which states that “the production, manufacture, fabrication, processing, preparation, storage, transportation, trafficking, sale, importation, supply and/or use of beta-agonist formulations that are not duly registered with and authorized by the Ministry are strictly forbidden.” This standard is of mandatory compliance and applies to all entities engaged in cattle farming, including slaughterhouses and establishments that produce, manufacture, fabricate,

¹⁷ J. L. Davis, G.W. Smith, R. E. Baynes, L.A Tell, A. I. Webb, and J.E. Riviere, “FARAD Digest. Update on drugs prohibited from extralabel use in food animals”, *Journal of the American Veterinary Medical Association*, 5 (2009), 529-530. <http://www.farad.org/publications/digests/092009ProhibitedDrugsUpdated.pdf> [Accessed 15 November, 2016].

¹⁸ Food and Agriculture Organization of the United Nations, “Clenbuterol. Maximum Residue Limits”, *Residues of some veterinary drugs in animals and foods. FAO Food and Nutrition Paper*, 41/9 (1997), <http://www.fao.org/docrep/W4601E/w4601e06.htm> [Accessed 15 November, 2016].

¹⁹ J. L. Davis *et al*, *ibid*.

process, prepare, store, transport, traffic, sell, import, supply, and/or use non-authorized beta-agonist formulations in animals, even when these are obtained free of charge.

In 2014, the Mexican government issued its latest guidelines to set criteria to determine MRL on products of animal origin.²⁰ The purpose of this agreement was to develop an effective and reliable toxic and contaminants residue control programme to participate in global and national trade. A national chart on MRL accompanied these guidelines. This supplementary document sets forth a zero-tolerance limit for clenbuterol on equine cattle (muscle, fat, liver and kidneys).²¹

The clenbuterol analysis done in this study reveals that none of the samples fulfilled these specifications. Figures 28 and 29 below illustrate the results. Figure 28 shows the amount of clenbuterol residue per positive sample taken from horses. As can be seen, all but two samples exceed even the MRL permitted by FAO for human consumption. The MRL is 0.2 µg/kg, meaning that only two positive samples do not exceed the limit.

²⁰ Secretaría de Gobernación, *Diario Oficial de la Federación*, “Acuerdo por el que se establecen los criterios para determinar los límites máximos de residuos tóxicos y contaminantes, de funcionamiento de métodos analíticos, el Programa Nacional de Control y Monitoreo de Residuos Tóxicos en los bienes de origen animal, recursos acuícolas y pesqueros, y Programa de Monitoreo de Residuos Tóxicos en animales, así como el módulo de consulta, los cuales se encuentran regulados por la Secretaría de Agricultura, Ganadería, Desarrollo Rural, Pesca y Alimentación”, 9 October, 2014.

²¹ Servicio Nacional de Sanidad, Inocuidad y Calidad Agroalimentaria de la Secretaría de Agricultura, Ganadería, Desarrollo Rural, Pesca y Alimentación, “Límites máximos de residuos tóxicos y contaminantes”, December 2014, <http://publico.senasica.gob.mx/?id=6351> [Accessed 15 November, 2016].

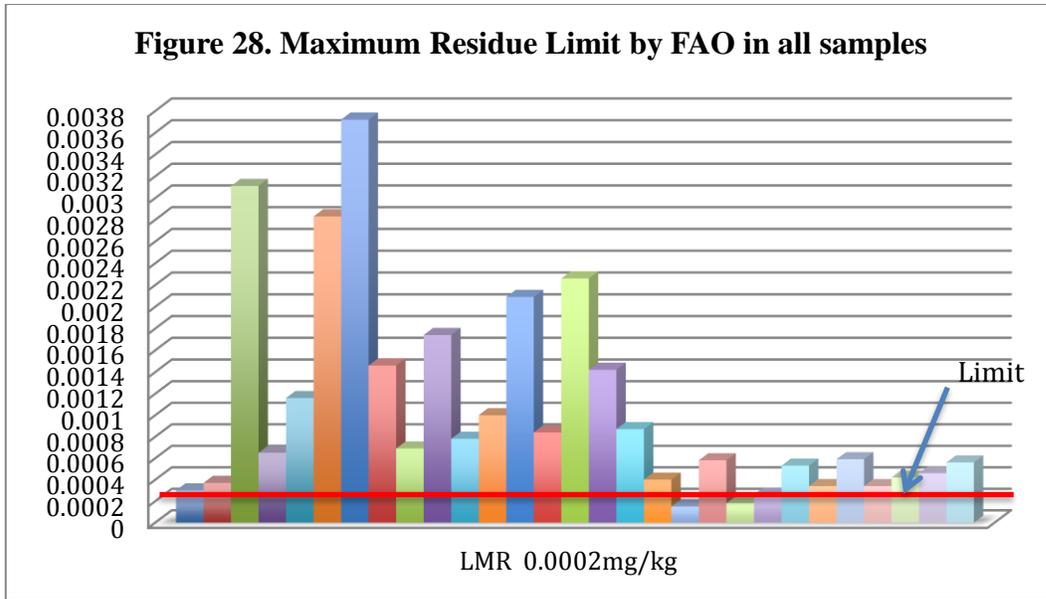
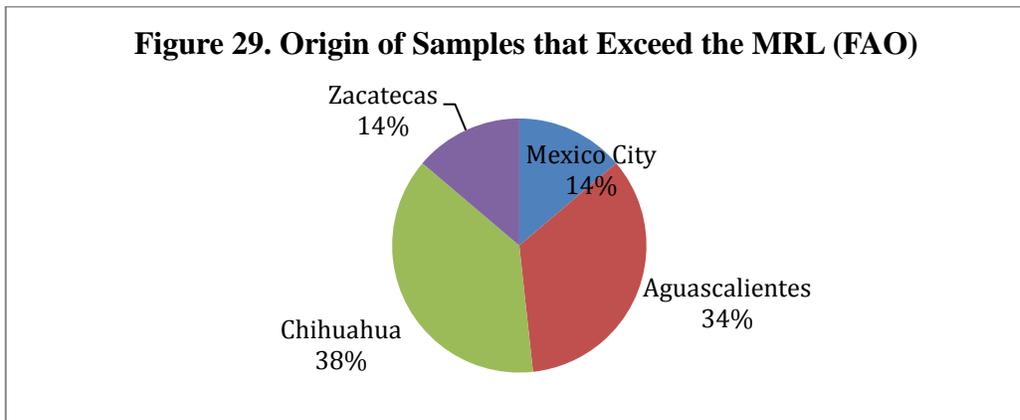


Figure 29 shows the percentage of samples that exceed the MRL (FAO) per city, i.e. of the samples that exceed the limit, 38% are from Chihuahua, 34% from Aguascalientes, 14% from Mexico City, and the remaining 14% from Zacatecas.



V. CONCLUSIONS

This study builds on earlier research and makes a modest contribution to the study of the substitution of species in meat products in developing countries in general, and in Mexico in particular – for which there is still a lack of information. Further research is needed and encouraged to gain more clarity on meat mislabeling practices in Mexico.

The results of the interviews conducted in the six different cities reveal an evident lack of interest in selling horsemeat among retailers. The same results were obtained with

regard to poor knowledge of horsemeat suppliers or where to buy this meat. This is due to several reasons, mainly:

1. The belief that offering equine meat intended for human consumption for sale is illegal.
2. The assumptions of the toxicity of horsemeat and the potential health risks it can generate through human consumption.
3. As in other countries, the association of horsemeat consumption with poverty and a low social status.

These reasons predominate in the cities surveyed in this study, even though in the northern states of the country, there is a wider knowledge that the sale of horsemeat is permitted. The study has also revealed that in most cities where samples were collected, except in San Vicente Chicoloapan, the use of horsemeat as a substitute for beef is a practice encountered in most types of establishments with the exception of supermarkets, and in all types of meats sampled.

A finding of 9.93% of samples testing positive for horsemeat demonstrates that horsemeat is effectively being commercialized in outlets, thus representing fraud to consumers. Additionally, the fact that 93.10% of selected samples exceeded the maximum residue limit for clenbuterol issued by the FAO and 100% exceeded the MRL according to the zero-tolerance limit provided by the Mexican regulations, demonstrates that Mexican consumers are being exposed to potential health risks.

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ANNEX 1. Cities, establishments and their localization in each city where meat has been purchased.

MEXICO CITY

North West	ZONE A	Mexico City
Establishment	Delegation	Place name
Market	Azcapotzalco	Laminadores
Market	Benito Juárez	La Postal
Street market	Cuauhtémoc	Santa María La Rivera
Street market	Gustavo A. Madero (GAM)	Politécnico
Street market	Benito Juárez	La Moderna
Supermarket	Azcapotzalco	Soriana Cuitláhuac
Supermarket	Cuauhtémoc	Wal-Mart Buenavista
Butcher shop	Azcapotzalco	Súper Carnicería La Dalia
Butcher shop	Benito Juárez	La Única

North East	ZONE B	Mexico City
Establishment	Delegation	Place name
Market	GAM	Rastro
Market	GAM	San Felipe de Jesús
Market	Venustiano Carranza	Merced
Street market	Venustiano Carranza	Aluminio
Street market	GAM	Guadalupe-Tepeyac
Supermarket	GAM	Chedraui Coyol
Supermarket	GAM	Bodega Comer. Mex. Sn. Juan A.
Butcher shop	GAM	Luis Antonio
Butcher shop	Venustiano Carranza	Refugio
Butcher shop	Iztacalco	La Chiquita

South West	ZONE C	Mexico City
Establishment	Delegation	Place name
Market	Magdalena Contreras	Cerro del Judío
Market	Álvaro Obregón	Olivar del Conde
Street market	Álvaro Obregón	Plateros
Street market	Magdalena Contreras	El Tanque
Supermarket	Álvaro Obregón	Bodega Aurrera Centenario
Supermarket	Álvaro Obregón	Wal-Mart Plateros
Supermarket	Álvaro Obregón	Mega Comercial Mexicana San Jerónimo
Butcher shop	Magdalena Contreras	Carnicería y Pollería

Butcher shop	Álvaro Obregón	La Súper
Butcher shop	Álvaro Obregón	La Gloria

South East	ZONE D	Mexico City
Establishment	Delegation	Place name
Market	Xochimilco	Mercado de Xochimilco
Market	Tláhuac	Mercado Central de Tláhuac
Market	Iztapalapa	Central de Abastos
Street market	Iztapalapa	Gpe. Del Moral/Escuadrón 201
Street market	Tlalpan	Villa Coapa
Street market	Xochimilco	Deportiva
Street market	Tláhuac	Del centro de Tláhuac
Supermarket	Coyoacán	Wal-Mart Villa Coapa
Supermarket	Benito Juárez	Mega Comer. Mex. Asturias
Butcher shop	Xochimilco	El Abuelo
Butcher shop	Tláhuac	Luis

ZACATECAS

NORTH	ZONE A-B	ZACATECAS
Establishment	Neighborhood, Area or Place	Place name
Market Jesús	Centro	Don Nacho
Market de la Plata	Boulevard de la Plata	Princesa
Market Jesús	Centro	Los Burros
Market	Estación FFCC	Gorditas Pachis
Market de la Plata	Boulevard de la Plata	Comidas Andrea
Supermarket	La Condesa	Soriana Mercado
Butcher shop	Alma Obrera	La Preferida
Market	La Condesa	Tacos Tal'Ivan
Butcher shop	La Condesa	Helguera
Butcher shop	La Condesa	La Ternerita
Butcher shop	La Condesa	Condesa
Butcher shop	La Condesa	La Rancherita
Supermarket	La Condesa	Soriana Súper
Butcher shop	Universidad	Carne Mart

SOUTH	ZONE C-D	ZACATECAS
Establishment	Neighborhood, Area or Place	Place name
Butcher shop	Abastos	El Cuñado
Market Guadalupe	Guadalupe	Antojos Zacatecanos Mary
Market Guadalupe	Guadalupe	San Francisco
Butcher shop	Guadalupe	Carne Mart
Butcher shop	Abastos	R - Villa
Butcher shop	Abastos	Jazmín

Street market	Abastos	Birria Julia
Street market	Abastos	Antojos Zacatecas
Street market	Abastos	Los Sopes
Street market	Abastos	Gorditas Mayra
Market Abastos	Abastos	Tacos Israel
Supermarket	Lomas del Campestre	Wal-Mart
Supermarket	Lomas del Campestre	Soriana Híper
Butcher shop	Lomas del Campestre	La Estrella
Butcher shop	Lomas del Campestre	Cortes San Francisco
Street market	Guadalupe	Carnicería
Street market	Guadalupe	Carnicería
Street market	Guadalupe	Carnicería

AGUASCALIENTES

NORTH	ZONE A-B	Aguascalientes
Establishment	Place name	
Supermarket	Comercial Mexicana	2do Anillo, Aguascalientes
Supermarket	Soriana Híper	
Market Terán	Carnicería Los Brothers	
Market Terán	Carnicería Puerta del Sol	
Supermarket	Bodega Aurrera, Guadalupe	
Others, 3er anillo, siglo XXI	Mrs. Pepe's Burguers	
Market Juárez	Antojitos Juárez	
Market Terán	Los Tacos	
Street market Cumbres	Antojitos Varios	
Market San Felipe	Carnicería Flamingos	
Street market San Felipe	Antojitos varios	
Others, Av. Convención 1914	Taquería convención	
Market San Felipe	Taquería san Felipe	
Butcher shop, Calle Libertad, Centro	Carnicería la Esperanza	

SOUTH	ZONE C-D	Aguascalientes
Establishment	Place name	
Market San Felipe	Carnicería Flamingos	
Others, Centro	Tacos Raúl	
Butcher shop Las cruces	Carnicería Olmos	
Butcher shop Las cruces	Carnicería Taurina VI	
Butcher shop Las cruces	Carnicería Picacho	

Butcher shop 3er anillo, siglo XXI	Carnicería La Esperanza
Market Purísima	Carnicería los Toños
Street market Purísima	Tacos
Market Purísima	Carnicería Obrador Soto
Market Purísima	Carnicería Manolos
Market Bonanza	Carnicería Bonanza, Real de Carnes
Street market, Las cruces	Tianguis
Supermarket, 3er anillo, siglo XXI	Súper Bara
Supermarket	Soriana Mercado
Supermarket	Bodega Aurrera
Supermarket	Comercial Mexicana
Supermarket	Chedraui
Market, Central de abastos	Súper carnicería San Juan
Supermarket	Wal-Mart Aguascalientes
Market de abastos	Carnicería Gonvi
Supermarket	H-E-B Aguascalientes
Street market centro comercial Agropecuario	Carnicería Carnes Supremas El recreo
Street market centro comercial Agropecuario	Carnicería Capricho (caballo)
Market Reforma	Obrador San Francisco
Street market	Gordas Purísima

CHIHUAHUA

NORTH	ZONE A-B	Chihuahua
Establishment	Place name	
Street market 2000	Tacos	
Street market 2000	Hamburguesas "Mike"	
Street market 2000	Tacos "Mike"	
Street market 2000	Tacos bistec	
Street market 2000	Burritos	
Street market 2000	Tacos bistec	
Street market 2000	Tacos	
Supermarket	Tacos "Doña Tuta"	
Supermarket	Burritos "Doña Tuta"	
Supermarket	Tacos "Chihua"	
Supermarket	Barbacoa "Chihua"	
Supermarket	Molida "S'mart"	
Supermarket	Bistec "S'mart"	
Street market Tarahumara	Tacos birria "Tacos y Lonches"	
Street market Tarahumara	Tacos "Tripitas"	
Street market Tarahumara	Carne "Brenda"	
Street market Tarahumara	Burritos	
Street market Tarahumara	Burritos	
Street market Tarahumara	Gordita Picadillo	

SOUTH	ZONE C-D	Chihuahua
Establishment	Place name	
Dairy and convenience store	Molida "La Nueva 28"	
Convenience store and butcher shop	Molida "Cuquita"	
Market Abastos	Burrito	
Market Abastos	"Beef Internacional"	
Hypermarket soriana	Tacos Barbacoa	
Market Abastos	"Carne Mart"	
	"Carnes y Derivados de Chihuahua"	
San Jorge	"Mini Súper López"	
Butcher shop and convenience store	"Abarrotes y Carnicería Ramírez"	
Butcher shop	Molida "Supercarnicería Las águilas"	
Cemetery San Jorge	Burritos "Los compadres"	
Cemetery San Jorge	Burritos	
Street market	Burritos "Mercado"	
Street market San Jorge	Tacos Barbacoa	
Street market San Jorge	Burrito	
Street market San Jorge	"Tacos y tripititas"	
Taco bar	"Estilo Chihua"	
Taco bar	"Av. Industrias"	
Taco bar	"Barbacoa Bruno"	
Taco bar	"Birrieria y Taquería Anita"	
Butcher shop	"Carne de Caballo"	
Butcher shop	"Carnicería Flores"	
Taco bar	"Las Brujas"	

Butcher shop	"Las Palomas"
Market Popular	"Antojitos Susy"
Cemetery San Jorge	"Los Compadres"

PACHUCA

North-South	Center	Pachuca
Establishment	Place name	
New market Benito Juárez	Tacos Soto	
Facing new market B. Juárez	Los Michoacanos	
Market Miguel Hidalgo	Carnicería Suprema	
Market Miguel Hidalgo	Tacos Julián	
Market Aquiles Serdán	Taquería El Tapatío 2	
Butcher shop	Distribuidora de Carnes Angus	
Others, Central de abastos	Taquería Aries	
Market, Central de abastos	Carnicería Vargas	
Butcher shop Mdo. Francisco I. Madero	Carnicería "La Surtidora"	
Butcher shop	Súper Carnicería "Lucy"	
Market, Central de abastos	Distribuidora "San Francisco"	
Others, Taco bar (José María Morelos y Ocampo)	Pollos Guerrero	
Others, Taco bar	Tacos "Sr. Limón"	
Others, Pastes (Leandro Valle)	Pastes Tuzo	
Market 1 de mayo	"La Favorita"	
Butcher shop	"La Única"	
Market de Barreteros	Cocina la Güerita	
Market de Barreteros	Carnicería Sagitario	
Market de Barreteros	Carnicería Don Beto	
Others, Pastes	Pastes Soto	

SAN VICENTE CHICOLOAPAN

North-South	Center	San Vicente Chicoloapan
Establishment	Place name	
Supermarket	Soriana	
Butcher shop	El Güero	
Street market Allende	Tacos	
Market Allende	Carnicería Leo 2	
Street market Ara 3	Tacos	
Butcher shop	Lupita	
Market Sare	Carnicería La Favorita 2	
Street market Ciudad Los Reyes 2	Tacos	
Street market Ciudad Los Reyes 2	Carnicería	
Supermarket	Bodega Aurrerá	

ANNEX II Photos of meat samples



Various samples refrigerated before being packed for transportation.



Various packed samples, labeled and ready to be sent to ITSON laboratory.



Various packed samples, labeled and ready to be sent to ITSON laboratory.