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Research Reports

Monitoring of Animal Products Entering East Kalimantan with Total Plate Count Test

Pemantauan Produk Hewan yang Masuk Kalimantan Timur dengan Uji Angka Lempeng Total

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ABSTRACT

Background: Quarantine measures on animal products that are transported between areas are prioritised at the place of export. It is necessary to establish a supervisory system through monitoring the entry of animal products. **Purpose:** Monitoring of animal products as food is carried out as an evaluation and consideration for further quarantine measures as well as for monitoring food safety aspects. **Method:** The test conducted on the samples taken was the Total Plate Count (TPC) test. A total of 32 chicken meat samples, 31 meat samples (1 lamb and chevon sample, 10 buffen samples, 29 beef samples), 23 chicken sausage samples, and 18 beef sausage samples. **Results:** A total of 21 samples (91.60%) of chicken sausage TPC test results in comply with SNI and and 2 samples (8.70%) TPC test results do not comply with SNI. Fifteen samples (83.33%) of beef sausage TPC test results in comply with SNI and 3 samples (16.67%) TPC test results do not comply with SNI. A total of 32 chicken meat samples, 1 lamb and chevon sample, 10 buffen meat samples, 29 beef samples the TPC test results were 100% in compliance with SNI 7388:2009. **Conclusion:** This can be caused by sanitation and hygiene of handling and storage, temperature on the conveyance when being transported and storage temperature, product packaging. There was a break when the unloading officer took a break. This condition causes the commodity to stay too long in the open container and the temperature becomes less stable. This is thought to be due to handling during distribution, instability of storage temperatures and the perishable nature of processed meat products.

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ABSTRAK

Latar Belakang: Tindakan karantina terhadap produk hewan yang diangkut antar wilayah diutamakan di tempat pengeluaran. Perlu dibangun sistem pengawasan melalui pemantauan masuknya produk hewan. **Tujuan:** Pemantauan produk hewan sebagai pangan dilakukan sebagai bahan evaluasi dan pertimbangan tindakan karantina selanjutnya serta pemantauan aspek keamanan pangan. **Metode:** Pengujian yang dilakukan terhadap sampel yang diambil adalah uji Angka Lempeng Total. Sebanyak 32 sampel daging (1 sampel daging domba dan kambing, 10 sampel daging kerbau, 29 sampel daging sapi), 23 sampel Sosis Ayam, dan 18 sampel Sosis Sapi. **Hasil:** Sebanyak 21 sampel (91,60%) sosis ayam hasil uji ALT sesuai SNI dan 2 sampel (8,70%) hasil uji ALT tidak sesuai SNI. Hasil uji ALT sosis sapi sebanyak 15 sampel (83,33%) sudah sesuai SNI dan 3 sampel (16,67%) hasil uji TPC tidak sesuai SNI. Sebanyak 32 sampel daging ayam, 1 sampel daging domba dan kambing, 10 sampel daging kerbau, 29 sampel daging sapi hasil uji ALT 100% memenuhi SN 7388 2009. **Kesimpulan:** Hal ini dapat disebabkan oleh sanitasi dan higienitas penanganan dan penyimpanan, suhu pada alat angkut pada saat diangkut dan suhu penyimpanan, kemasan produk. Terjadi jeda saat petugas bongkar muat istirahat. Kondisi ini menyebabkan komoditas terlalu lama berada dalam wadah terbuka dan suhu menjadi kurang stabil. Hal ini diduga disebabkan oleh penanganan saat pendistribusian, ketidakstabilan suhu penyimpanan dan sifat produk daging olahan yang mudah rusak.

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Kata kunci: Angka Lempeng Total; Karantina; Pemantauan; Produk Hewan

INTRODUCTION

Currently, the Indonesian government has issued policies contained in Economic Package XI related to Indonesia Single Risk Management (ISRM) (2016) and Economic Package XV (2017) related to the Simplification of Commerce (export-import) in order to accelerate the flow of goods and services at the port and to reduce dwelling time. This policy also requires quarantine officials in carrying out quarantine measures to be able to facilitate and accelerate the flow of goods at the place of entry and exit. The Indonesian Quarantine Agency through the Technical Implementation Unit (UPT) of the Animal, Fish and Plant Quarantine Center (BBKHIT) East Kalimantan plays an active role in efforts to prevent the entry, spread and release of Quarantine Animal Pests and Diseases (HPHK). Animal, Fish and Plant Quarantine Center (BBKHIT) East Kalimantan carries out the main task functions including the implementation of 8P "Pemeriksaan, Pengasingan, Pengamatan, Perlakuan, Penahanan, Penolakan, Pemusnahan, dan Pembebasan" of HPHK, one of which is animal products in comply with Law No. 21 of 2019.

Quarantine measures for animal products transported between areas within the Republic of Indonesia are primarily carried out at the exit point. At the entry point, quarantine officers will inspect documentation and the integrity of the packaging. Animal products in transit must be guaranteed to remain in good condition during the logistics process and that there is no change in quality. However, it is also necessary to consider the acceleration of goods and services flow in ports and dwelling time. Therefore, it is necessary to build a monitoring system for supervision. The monitoring carried out is also related to the problem of microbial contamination in animal products, such as beef, mutton, beef sausage, chicken sausage and others. The problem of contamination is because there are obstacles in maintaining the freshness and quality of animal products. Meat, for example, can easily deteriorate during processing and storage (Kana *et al.*, 2021). The importance of monitoring can also be seen from the consumer side, where they are very concerned about the physical signs of fresh animal products, such as meat. Therefore, it is very important to monitor raw animal products (Torres *et al.*, 2013). The result of microbial contamination that was not detected can certainly lead to further problems, such as spoilage in animal products. Most of the meat and meat products spoil every year. Approximately 3.5 billion kg of poultry meat is wasted at the consumer, meat producer and eatery level. This very high loss results in significant economic and environmental impacts (Ajaykumar, 2020).

Monitoring is carried out on food animal products that are crossed between islands. In supporting the implementation of monitoring, the role of the Animal, Fish and Plant Quarantine Center (BBKHIT) East Kalimantan is very important, especially in conducting TPC testing of animal product samples that are passed into the East Kalimantan region. The purpose of this research through TPC testing is to obtain monitoring data on animal products entering the East Kalimantan region. In addition, the results of this study also aim to be used as material for evaluation and consideration of quarantine measures, as well as animal quarantine service policies in the prevention of Quarantine Animal Pests and Diseases (HPHK) and animal biosafety. Furthermore, to support quarantine supervision in compliance with food safety aspects.

MATERIALS and METHODS

Methods

Testing was carried out on samples of carrier media in the form of animal products entering Kaltim Kariangau Terminal Port from March to October 2019. The product samples taken are animal products entering East Kalimantan. The frequency of sampling refers to the Decree of the Head of the Agricultural Quarantine Agency Number: 2464/Kpts/Kr.120/K/11/2018 concerning Guidelines for Monitoring Animal Origin Materials and Products of Animal Origin Materials. Tests carried out on samples are the Total Plate Count (TPC) to count bacterial colonies grown on agar media.

Experimental Design

Samples taken in the context of monitoring animal products include chicken meat, lamb, chevon, buffen, beef, chicken sausage, and beef sausage. These samples are animal quarantine commodities that are passed into East Kalimantan through the Kaltim Kariangau Terminal Port. The test conducted was the TPC test at the East Kalimantan BBKHIT Laboratory. TPC testing is a total plate test that functions to calculate the number of microbes contained in a product by counting bacterial colonies grown on agar media. How to calculate the number of microbes contained in a product that grows on agar media at a specified temperature and incubation time (SNI 2897:2008). Total Plate Count is useful to indicate quality, shelf life/half-life, contamination and hygienic status during the production process.

Testing Procedure

Solvent Preparation Stage

Weigh 25.5 grams of Buffer Peptone Water (BPW) and dissolve it in 1000 ml of distilled water. Next, sterilise the BPW solution in an autoclave at 121°C for 15 minutes.

Sample Preparation Stage

Weigh 25 grams of solid and semi-solid samples and place them in sterile plastic containers. Add 225 ml of sterile BPW solution to the plastic containers containing the samples and homogenise with a stomacher for 1-2 minutes. This will result in a solution with a dilution of 10-1. Prepare serial decimal dilutions as needed from solutions 10-2, 10-3, 10-4, and 10-5 with BPW solution.

Analysis Stage

The pour plate method was used to culture the samples. One millilitre of each decimal dilution series was pipetted into a sterile petri dish in duplicate.The autoclaved PCA was cooled to a temperature of 45°±1°C and 15-20 ml was aseptically added to each petri dish containing the sample solution. To ensure proper mixing of the sample solution and media,

rotate the Petri dish clockwise several times. Allow the media to harden before proceeding. Place the dish upside down and incubate it in an incubator at 34-36ºC for 24-48 hours. After incubation, count the plates that have 25-250 colonies.

Analysis Data

Data obtained from the TPC test results were subjected to descriptive statistical analysis. This data is derived from total samples taken from animal products entering through Kaltim Kariangau Terminal. The data acquired is then used to see samples that comply or do not comply with the maximum limit of microbial contamination in food (SNI 7388, 2009).

RESULTS

The samples taken were 114 animal products. The results of testing the number of microbial contaminants with the TPC test on 114 samples of animal products that were transshipped through the Kaltim Kariangau Terminal Port are presented in Table 1 and Table 2.

Total Plate Count Result

Tabel 1. Percentage of Meat TPC Test Results.

No.	Sample Type	Microorganism Contamination Limit	Percentage of TPC Test Result (%)
1	Chicken Meat	Compliant with SNI	100
	(32 samples)	Not compliant with SNI	0
2	Lamb	Compliant with SNI	100
	(1 Sample)	Not compliant with SNI	0
	Chevon	Compliant with SNI	100
	(1 Sample)	Not compliant with SNI	0
	Buffen	Compliant with SNI	100
	(10 Samples)	Not compliant with SNI	0
	Beef	Compliant with SNI	100
	(29 Samples)	Not compliant with SNI	0

Tabel 2. Percentage of Sausage TPC Test Results.

No.	Sample Type	Microorganism Contamination Limit	Percentage of TPC Test Result (%)
1	Chicken Sausage	Compliant with SNI	91.30
	(23 Samples)	Not compliant with SNI	8.70
2	Beef Sausage	Compliant with SNI	83.33
	(18 Samples)	Not compliant with SNI	16.67

The testing was conducted on chicken meat, lamb, chevon, buffen, beef, chicken sausage, and beef sausage that had transhipped through Kaltim Kariangau Terminal. The samples were cultured using the pour plate method in the laboratory of BBKHIT East Kalimantan.

Based on Table 1 of the TPC Test Results, 32 samples of chicken meat TPC test results 100% in comply with SNI or still below the maximum limit of microbial contamination, 1 sample of lamb and chevon TPC test results 100% in comply with SNI, 10 buffen samples TPC test results 100% in comply with SNI, 29 beef samples TPC test results 100% in comply with SNI, 21 Samples (91.30%) chicken sausage TPC test results in comply with SNI and 2 samples (8.70%) TPC test results do not comply with SNI, namely 8.75 x 105 cfu/g and 1.29 x 105 cfu/g, 15 samples (83.33%) beef sausage TPC test results in comply with SNI and 3 samples (16.67%) TPC test results do not comply with SNI, namely 7.1 x 106 cfu/g, 2.2 x 105 cfu/g, and 2.1 x 105 cfu/g respectively.

DISCUSSION

Meat TPC Result

The TPC test results were compared with SNI 7388:2009 regarding the Maximum Limit of Microbial Contamination in food. According to the standard, the maximum limit for fresh and frozen meat is $1x10^6$ colonies/g, while for processed meat products, it is $1x10^5$ colonies/g. According to SNI (2000), the Maximum Limit of Microbial Contamination (BMCM) is the maximum number of microbes (cfu/gram or cfu/ml) that are permitted or recommended to be acceptable in foodstuffs of animal origin. The classification of BMCM in foodstuffs of animal origin is classified into one quality level. The maximum limit of microbial contamination in food in the TPC test for fresh and frozen chicken meat, fresh and frozen meat is $1x10^6$ colonies/g, with processed meat and processed chicken meat (meatballs, sausages, nuggets, burgers) is $1x10^5$ colonies/g (SNI 7388, 2009). There are 2 samples chicken sausage and 3 beef sausage samples that do not comply with SNI 7388:2009.

There are several factors that cause the product test results to not comply with SNI or be above the maximum threshold for microbial contamination, presumably due to sanitation and hygiene of handling and storage, temperature on the conveyance when being transported and storage temperature in the warehouse, and product packaging. From the results of the inspection and review of the animal product warehouse location, it is suspected that this occurred because during unloading from the container to the warehouse, there was a break when the unloading officer took a break. This condition causes the commodity to stay too long in the open container and the temperature becomes less stable. In addition, it is also due to the condition of the cold room which sometimes experiences disturbances so that the temperature is sometimes less stable. Fluctuating temperatures can increase microbial growth in animal products, resulting in unfavourable effects. During three meals, the number of doors opened can reach 40 to 50 times, which can also contribute to unstable temperatures. Additionally, the thawing process can cause the temperature inside the freezer to rise, which can have adverse effects on meat stored in cold storage (Zhao *et al.*, 2017).

Temperature fluctuations can occur during various food storage durations due to factors such as frequent opening of the door and defrosting.These factors cause air exchange inside and outside the refrigerator and physical changes, resulting in frequent temperature fluctuations in cold storage (Khan, 2014). Soeparno (1992) categorised factors influencing the growth of microorganisms in meat into intrinsic and extrinsic factors. Intrinsic factors include the nutritional composition of the meat, water activity, pH level, oxidation-reduction potential, and the presence or absence of

barrier substances. These factors are directly related to the inherent characteristics of the meat itself and can either promote or inhibit microbial growth by providing suitable or unsuitable environments for their proliferation. Extrinsic factors, on the other hand, include temperature, relative humidity, oxygen availability, and the physical condition of the meat. The rate of microbial growth in meat is significantly influenced by environmental temperature and humidity, as well as by oxygen availability and meat properties such as surface area and particle size. It is important to understand these factors in order to effectively control microbial growth in meat products and ensure food safety and quality.

Sausage TPC Result

Other factors that can cause microbial contamination in food products of animal origin are the handling of food products that are not in accordance with hygiene requirements, such as raw materials, equipment used, processing, serving, and storage that are not hygienic (Cahya *et al.*, 2019). Furthermore, environmental factors such as pH, temperature, and oxygen can also cause microbial growth (Nurmila and Kusdiyantini, 2018). Sanitation and hygiene relate to the handling of animal products when they are distributed from containers to storage warehouses. Poor hygiene and sanitation practices in product handling can result in microorganism contamination. Microorganism can multiply easily in animal products such as sausages, because sausages are processed meat products that are perishable, the nutritional content in hem can be utilised by microorganisms to live. Efforts need to be made to reduce contamination in food or food products, namely maintaining cleanliness and sanitation, both in the selection of raw materials to storage (Tama *et al.*, 2023). Microbial growth in unwanted foodstuffs can be found in the form of food damage and diseases that arise from consuming food products contaminated with pathogenic microbes (foodborne disease) (Yusuf *et al.*, 2016).

Storage temperature also affects the quality of animal products. According to Agricultural Quarantine Agency Decision Number: 2205/KPTS/KR.120/K/12/2017 Regarding Guidelines for the Entry and Exit of Animal Products Within the Territory of the Republic of Indonesia, the recommended storage temperatures for animal products for consumption transported using special transport equipment (refrigerated containers/reefer containers) are as follows: frozen animal products, including frozen meat, sausages, and nuggets, should be stored at temperatures ranging from -18°C to -22°C; and fresh chilled animal products, including fresh meat and cheese, should be stored at temperatures ranging from 2°C to 6°C. Temperature checks are part of the physical inspection carried out by animal quarantine officers on animal products. In addition to the physical inspection of animal products and packaging, as outlined in Agricultural Quarantine Agency Decision Number: 2205/KPTS/KR.120/K/12/2017, the examination of temperature is crucial. Product packaging is also essential to prevent contamination by microorganisms, as per Indonesian National Standard (SNI) 3820:2015 for meat sausages, ensuring that products are packed securely to prevent deviations or damage during storage and transportation. In Usmiati's (2010) study, meat deterioration due to microbial activity is primarily indicated by the proliferation of spoilage microorganisms, which cause the formation of mucus and metabolic by-products. Additionally, changes in color are also visible signs of meat spoilage, often accompanied by shifts in hue, indicating biochemical changes within the meat matrix. Furthermore, the presence of unpleasant smells, caused by the breakdown of proteins and the creation of malodorous substances such as ammonia, hydrogen sulfide (H_2S), and mercaptans, indicates significant microbial spoilage. Alterations in flavour, which are described as sourness and bitterness, are the result of the metabolic processes of acid-forming bacteria and the generation of bitter compounds during microbial growth. The deterioration of meat quality is contributed to by the onset of rancidity, which results from the breakdown or oxidation of meat fat. These observable indicators are crucial parameters for assessing meat quality and safety, emphasising the importance of microbial control measures in meat preservation practices.

Meat sausage is a product made from mashed meat with or without the addition of other food ingredients and permitted food additives and inserted into sausage casings with or without a cooking process (SNI 3829:2015). Meat for sausages can come from beef, buffen, chevon, lamb, pork, poultry or other farm animals. Sausage is a processed meat product that is susceptible to bacterial contamination and has the potential to cause food poisoning of animal origin (Ed-dra *et al.*, 2017). The sources of contamination in sausages include cooking and serving utensils, raw materials, improper timing, and temperature during food production, as well as personnel hygiene. Additionally, bacterial contamination in sausages is influenced by several factors, such as the sales location and the type of sausage. It is important to implement stringent quality control throughout the entire chain of production for animal-origin food (Ed-dra *et al.*, 2017).

The risk of contamination in animal products can be caused by the presence of bacteria. The contamination that occurs cannot be seen physically, but laboratory tests are required. In markets or supermarkets, we cannot know whether the bacterial colonies in animal products are still within the threshold or not. Some studies have shown the presence of contamination in pagan products of animal origin. The results of the study by Rehman *et al.* (2022) previously showed that chicken meat samples showed contamination with the pathogenic bacteria *E. coli* (34%), *Salmonella sp.* (28%), *Staphylococcus sp.* (25%), *Shigella sp.* (8%), *Enterobacter sp.* (2%) and *Bacillus sp.* (3%). Chevon samples *E. coli* (32%), *Salmonella sp.* (32%), *Staphylococcus sp.* (12%), *Shigella sp.* (12%), *Enterobacter sp.* (9%) and *Bacillus sp.* (3%). In beef samples *E. coli* (39%), *Salmonella sp.* (30%), *Staphylococcus sp.* (18%) and *Enterobacter sp.* (8%) and *Shigella sp.* (5%). Another study showed an increasing trend of dangerous food contaminants in various local foods. This microbial contamination, such as in chicken, beef and raw milk samples found in retail markets.

This problem is quite serious, with the total contamination of food samples and infected workers being 48.37%. Food samples were also found to contain *Salmonella spp.* (19%), *E. coli* (O157:H7) (8%) and 1.25% *Listeria monocytogenes*. In addition to bacteria, this contamination was also found to be caused by viruses. On further testing, 5% of the food samples were found to be contaminated with at least two viruses. The results of this study encourage the adoption of proper food hygiene practices to reduce the incidence of foodborne illness (Samad *et al*., 2018).

Microbial contamination that continues to grow in animal products has the risk of spoilage in these products. This condition is dangerous for consumers as it can lead to the entry of pathogens originating from animal products to humans.Contamination or the presence of pathogens in food of animal origin is a threat not only to Indonesia but to the whole world. Contamination of meat is also a gateway to zoonoses. Pathogens present in meat cause gastrointestinal infections, including diarrhoea and haemolytic uremic syndrome (Bantawa *et al*., 2018). Meat spoilage is a complex mechanism involving both biological and chemical reactions that result in meat products that are undesirable for human consumption. It results from the growth and feeding of various microorganisms on meat, leading to excessive release of metabolites or spoilage by enzymatic action (Casaburi *et al*., 2015). Only a small percentage of microorganisms that grow on meat can spoil the product through their metabolic activities. Microbial metabolism involves the consumption of metabolites and the production of new ones. For instance, when lactic acid bacteria (LAB) interact with carbon sources in meat, they produce lactic acid (Pothakos *et al*., 2015). This can affect meat texture by inducing protein precipitation and microbial protection due to the properties of lactic acid. The decrease in pH resulting from this process has an antimicrobial effect and produces a sour flavour (Zagorec, 2017). Microbial biotic and abiotic interactions in the meat substrate can occur during storage (Zhang *et al*., 2015). The microbial composition of meat can be affected by several factors. These factors relate to temperature control during slaughter, processing and distribution. Other factors relate to preservation techniques and type of packaging. Handling and storage by the consumer are also a factor in the microbial composition of meat. Hygiene during slaughter, evisceration and meat processing is also important (Kana *et al*., 2021). This is critical to evaluate animal products that are on the market in order to preserve food safety, particularly for consumers.

In response to this problem, quarantine measures for the entry of animal products need to be built with a monitoring system. Monitoring of animal products as food is carried out in order to supervise food safety aspects. The test of animal products that are shipped to Kaltim Kariangau Terminal Port with the TPC test is one of the monitoring measures for food safety. This is because according to SNI 2897 (2008) concerning Testing Methods for Microbial Contamination in meat, eggs, milk, and their preparations, TPC can show the number of microbes in a product by growing bacterial colonies grown on agar media. In addition, according to SNI 7388 (2009) the limit of microbial contamination in food, TPC can indicate food quality, storage period, contamination, and hygienic status during the production process. In addition to monitoring the animal products, there is a need for continuous evaluation of the temperature both during the transport process in the container and in storage. According to Agricultural Quarantine Agency Decision Number 2205 of 201, frozen animal products must be stored within the temperature range of -18°C to -22°C. Similarly, fresh chilled animal products should be stored between 2°C to 6°C. Low temperature is crucial in the storage of animal products as it can effectively control the growth of microorganisms and the effect of enzymes, thereby slowing down the rate of meat spoilage (Deng *et al*., 2020). Consistent storage and monitoring can prevent excessive microbial contamination in animal products. The application of low temperature is efficient in maintaining meat quality and extending shelf life, particularly during long-distance transportation of animal products, such as from Java to East Kalimantan.

Monitoring should be conducted on the packaging of animal products during transportation to ensure that it is properly sealed, undamaged, and does not pose a risk of microbial contamination to the products. Technologies to extend the shelf life of meat include vacuum packaging (ÇEt˙Inkaya, 2015), modified atmosphere packaging, and food preservatives (Silva *et al*., 2016). Vacuum packaging is a cost-effective way to extend the shelf life of meat. To continue reporting animal product commodities entering the East Kalimantan region, the support of business owners is necessary. Additionally, it is crucial for business owners and those in charge of transport equipment to maintain stable temperatures. Continuous monitoring by Animal, Fish and Plant Quarantine Center (BBKHIT) East Kalimantan can minimize the risk of microbial contamination in animal products. To ensure the quality and safety of food entering the East Kalimantan region, it is crucial to have the support of business owners in reporting all animal product commodities. Monitoring tests on animal products can be implemented to evaluate their quality and safety. This information can then be used to determine the need for further quarantine measures and support quarantine supervision in fulfilling

CONCLUSION

Total Plate Count testing of animal product samples that entered through Kaltim Kariangau Terminal Port obtained the following results, 95.61% of samples were within the normal range or below the limit of microbial contamination in food and 4.39% of samples exceeded the limit of microbial contamination in food found in beef and chicken sausages. This is thought to be due to handling during distribution, instability of storage temperatures and the perishable nature of processed meat products.It is necessary to carry out routine montoring of animal products to improve quarantine duties in preventing HPHK and support supervision of food safety aspects.

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CONFLICT of INTEREST

The author declares that the research was conducted in the absence of any commercial or financial. The authors have no conflict of interest in the results of the research conducted. The results of this study are a policy step for the supervision and prevention of HPHK, as well as related to the security of food of animal origin. relationships that could be construed as a potential conflict of interest.

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ETHICAL APPROVAL

This research activity did not require ethical approval.

AUTHORS' CONTRIBUTIONS

FR, RN: Conceptualization, FR, SSL : Design of study Methodology, DR, DU : Validation, Sample collection and analysis, DPS: Editing Manuscript, All authors have read, reviewed, and approved the final manuscript.

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