Therapeutic importance of timely immunophenotyping of breast cancer in a resource-constrained setting: a retrospective hospital-based cohort study

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Abstract

Background. The Mexican Consensus of 2015 on diagnosis and treatment of breast cancer proposes the use of surrogate classification based on immunohistochemistry (IHC). In this study, we assessed the therapeutic value of this consensus recommendation, in the absence of gene expression data, as a tool for providing opportune and precise care.

Methods and Findings. In this retrospective hospital-based cohort study, we reviewed medical records of all suspected breast cancer patients in 2014 treated at the General Regional Hospital No. 1, of the Mexican Institute of Social Security (IMSS), located in Charo, Michoacan (western Mexico). Then, we followed the medical history of those patients with IHC testing until March 2017. The trajectory of patients was recorded to discover common patterns, and turnaround time for IHC testing. There were 402 suspected breast cancer patients in 2014, of which 30 had been tested for some IHC biomarkers (ER, PR, HER2). The surrogate subtyping allowed doctors to adjust (56.7 %) or confirm (43.3 %) the initial therapeutic regimen. Opportune IHC testing was found to be beneficial when it was available before or during the first rounds of chemotherapy.

Conclusions. The use of opportune immunohistochemistry provides improved outcomes for breast cancer patients. This result has the potential to become a roadmap for the implementation of precision medicine in Mexico.

Keywords: Breast Neoplasms; Immunohistochemistry; Tumor Biomarkers; Medical Records; Clinical Coding; Mexico

Abbreviations: IMSS, Mexican Institute of Social Security; HGR1, General Regional Hospital No. 1 in Charo, Michoacan, Mexico.
1. Introduction

Molecular subtyping for breast cancer tumors based on gene expression patterns have been previously recognized to have prognostic value in the management and treatment of breast cancer patients [1]. During the last decade, there have been several commercially accepted multi-gene prognostic tests. Genomic prognostic tests have been associated with decreased adjuvant chemotherapy use among patients, and better outcomes with cost-effective treatments [2]. However, the availability and expense of these genomic tests remain largely prohibitive for constrain-resourced hospital settings, regardless of country income. For example, in the United States, the use of the 21-gene recurrence score Oncotype DX has increased significantly over the last decade due to insurance coverage, but it is estimated that only a quarter of eligible patients have been tested [3].

In resource-constrained settings, the use of surrogate immunohistochemistry (IHC)-based breast cancer subtypes presents as an alternative to molecular subtyping when there is no access to gene expression data or high-throughput sequencing data. Despite the lowering costs of genomic tests, these technologies are only available to researchers in select locations in the world, and they are inaccessible to most patients worldwide. Alternatively, a panel of three IHC markers—estrogen receptor (ER), progesterone receptor (PR), and human epidermal growth factor receptor 2 (HER2)—demonstrated its predictive value for chemotherapy response in breast cancer [4]. There are other IHC markers that could be included for the prediction of treatment response and prognosis; however, further studies are needed to select an optimized surrogate panel [5].

The 2015 Mexican Consensus on diagnosis and treatment of breast cancer [6] is the main working document for oncologists in the country. It recommends routine use of a 3-marker panel
(ER, PR, HER2) IHC approximation to identify four intrinsic breast cancer phenotypes (luminal A, luminal B, HER-2, and triple negative). In the Mexican population, the estimated frequency of ER/PR positive is 60%, HER2 positive 20.4%, and triple negative is 23.1% [7,8]. There is a troubling onset of disease among younger breast cancer patients in Mexico (aged <40), with a high prevalence of triple-negative breast cancers [9]. Furthermore, there is an increased mortality trend associated with breast cancer in the country [10].

The Mexican clinical practice guidelines for screening, management, and treatment of breast cancer [11-13] also recommends the use of IHC testing as part of the histopathology studies. Despite these recommendations, the widespread use of genetic testing in Mexico remains elusive due to constrained resources. However, IHC testing is widely available through hospital-owned pathology laboratories, as well as through outsourced commercial laboratories across the country.

In this study, we aimed to assess the timeliness of surrogate IHC biomarker testing in the context of a resource-constrained hospital setting. Evaluating the therapeutic value for patients is of critical importance, since the demand for more IHC testing will create new logistical challenges in the healthcare system. The impact of acceptable or critical time delays is ultimately not well defined, and it would be of great importance for hospital administrators to have a reference of the value of timely IHC testing. As precision medicine continues to provide better prognostic biomarkers, translating these benefits into clinical practice increasingly requires a time-efficient approach from the entire healthcare system.
2. Methods

Ethical approval

This research was reviewed and approved by the Mexican Institute of Social Security (IMSS)’s National Scientific Research Committee and its National Bioethics Committee, under protocol number R-2017-785-010, and locally by IMSS’ Local Research and Ethics Committee, under specific protocol number R-2017-1602-14 HGR1 Charo, Michoacan. Stanford’s Institutional Review Board provided a non-human subject determination under eProtocol: 40704. Researchers only received access to non-identifiable data under the rigorous considerations of the applicable Mexican bylaws on health research, patient privacy, and electronic medical records. Consent was not required.

Hospital setting

The Mexican Institute of Social Security is a hybrid single-payer system with an integrated network of hospitals nationwide. In Michoacan (a state located in western Mexico), the General Regional Hospital No. 1 (HGR1) is the designated secondary-care facility for the IMSS-insured population, estimated at 1,288,695 people (28% of the state’s population), out of 4,584,471 according to the 2015 census [14].

This hospital is in Charo, a suburban community adjacent to the Michoacan capital (Morelia), and it was built in 2012 to replace the old general hospital. HGR1 is the reference secondary-care facility, which serves as reference for seven General Zone Hospitals and 45 Family Medicine Units within the state of Michoacan. HGR1 has an oncology unit with a full complement of fixed staff and facilities available to all patients. It also offers pathology services, diagnostic imaging, and therapeutic capabilities with access to all approved drugs. Patients with
breast cancer who require chemotherapy are sent to the outpatient medical unit, and patients with breast cancer who require radiation therapy are provided the service through an outsourced private service in the same city. The personnel include gynecologists, medical oncologist, surgical oncologists with significant breast cancer training, adequate numbers of nursing and pharmacy staff, surgeons with significant training, and cancer pathologists. A self-assessment of the services and facilities provided by these hospitals is S1 Text, which is based upon a modified version of the oncological service self-assessment levels for low- and middle-income countries developed by The International Society of Pediatric Oncology (SIOP) committee on Pediatric Oncology in Developing Countries (PODC) [15].

Patients

The study population for this research was all cumulative breast cancer patients seen in 2014 by medical staff at the IMSS’s General Regional Hospital No 1 in Charo, Michoacan. The institution’s breast cancer census was used to select patients that were seen in 2014 at HGR1. To avoid selection bias in our study, we did not restrict the selection of patients to members of the female sex or any age group. The inclusion criteria can be seen in Figure 1.
A patient with suspected breast cancer is referred from a primary-care facility to HGR1 after being examined by his/her family physicians. If the physician suspects the existence of a breast lesion, the patient undergoes an imaging assessment using the breast imaging-reporting and data system (BIRADS) score. Upon arriving at HGR1, the patient was either seen by the breast cancer clinic (BIRADS 0 and 3) for further imaging studies and/or fine-needle biopsy assessments, or directly referred to the medical oncology service (BIRADS 4, 5 and 6), at which point they would undergo surgical biopsies for pathology analysis. The gold-standard diagnosis of breast cancer would be provided by a board-certified pathologist, who would assign one of the C50 codes (malignant neoplasms of breast) from the international code of diseases version 10 (ICD10).

We reviewed the medical records (both electronic and paper) of all breast cancer patients at HGR1, and selected those whose biopsies had also been tested with IHC to detect ER, PR, HER2 antibodies; then, we followed the medical histories of those patients, ending with March
2017, making note of multiple medical visits to the breast cancer clinic, medical oncology service, and/or surgical oncology service. From their medical records, we extracted information about IHC testing (antibody ordered, date of ordering, date of results being obtained), chemotherapy and hormonal drugs administered, and radiation sessions and surgical procedures undertaken.

Study design

This was a retrospective hospital-based cohort study, using medical records collected routinely as part of clinical care. The objective was to understand the therapeutic impact on breast cancer patients of the time taken to test surrogate immunohistochemistry biomarkers (ER, PR, HER2) in a resource-constrained hospital in western Mexico. We were interested in reporting the frequency of treatment selection or treatment adjustment frequencies depending on breast cancer subtyping and the turnaround times for surrogate immunohistochemistry biomarker testing. The analysis included timeline trajectories and association rules. The STROBE checklist is provided in S2 Text. All analyses were performed in R version 3.3.2.

Timeline trajectories. We explored the paper- and electronic-based medical records for all patients with IHC testing, from the patient’s first day at the hospital to the last follow-up pertaining to this study (occurring before March 2017). First, author MFRM manually reviewed the patient’s medical records and curated a set of possible events (transactions) that occurred to the patients of this study, annotating the time points for each event for each patient. Then, author ALP annotated those events with standardized coding from the Unified Medical Language System (UMLS). When disagreement occurred, both authors discussed the translation to assign a UMLS code to the medical records in Spanish. Finally, similar events were grouped together in
fewer categories. The UMLS codes, UMLS descriptions, Spanish description, and groupings can be seen in Table 1.

Table 1. List of medical events with UMLS codes and descriptions in English and Spanish.

| UMLS code   | UMLS description            | Description in Spanish                                      |
|-------------|------------------------------|------------------------------------------------------------|
| Visit       |                              |                                                            |
| C0008952    | Clinic Visits                | *Atención inicial en la unidad de medicina familiar (primer nivel de atención).* |
| C2153644    | Visit for: gynecological exam| *Atención inicial en la consulta de ginecología (segundo nivel de atención).* |
| Oncology    |                              |                                                            |
| C1620996    | Oncology; primary focus of visit; work-up, evaluation, or staging at the time of cancer diagnosis or recurrence (for use in a medicare-approved demonstration project) | *Atención inicial en la consulta de oncología médica.* |
| C1617848    | Oncology; primary focus of visit; expectant management of patient with evidence of cancer for whom no cancer-directed therapy is being administered or arranged at present; cancer-directed therapy might be considered in the future (for use in a medicare-approved demonstration project) | *Atención subsecuente en la consulta de oncología médica.* |
| Request lab |                              |                                                            |
| C2186763    | Request lab results from pathology | *Solicitud de estudios de patología.*                   |
| C2186756    | Request lab results from hematology | *Solicitud de estudios básicos de laboratorio clínico.*        |
| C2186777    | Request lab results from x-ray | *Solicitud de estudios de imagen (tele de tórax, ultrasonido, mastografía).* |
| C2186774    | Request lab results from CT   | *Realización de estudios especiales de imagen (tomografía).* |
| C2186775    | Request lab results from MRI  | *Realización de estudios especiales de imagen (resonancia magnética).* |
| Biopsy / Pathology |                        |                                                            |
| C0177666    | Needle biopsy of breast      | *Realización de biopsia con aguja fina o gruesa.*            |
| C0585992    | Surgical biopsy of breast    | *Realización de biopsia por escisión quirúrgica.*            |
| C0807321    | Pathology report             | *Reporte de estudio histopatológico.*                        |
| Chemotherapy|                              |                                                            |
| C0086965    | Selection for Treatment      | *Selección de tipo de tratamiento (quimioterapia, radioterapia, cirugía).* |
| C4302504    | Chemotherapy started         | *Inicio de quimioterapia.*                                  |
| Surgery / Radiation |                      |                                                            |
| C0436382    | Radiotherapy started         | *Inicio de radioterapia.*                                   |
Missing data. Missing data was assumed to be non-missing at random (NMAR), and the
events from Table 1 were considered missing for a specific reason. For example, if the code
C2186775 (request lab result from MRI) was missing from a patient’s medical record, we left
that value unassigned, since it was assumed that an MRI was not requested for that patient. There
is a small chance that this assumption might be violated (and introduce some bias) if the
physician did request the exam/procedure but forgot to write it down in the medical record.
However, we did not impute any missing data, since that might be a larger source of bias for this
EHR-based data.

Outliers. We assessed the time-to-event distribution for each event in Table 1, and
reported the mean time using boxplots with whiskers. The outliers were identified using the 1.5
interquartile rule, but were not removed from the downstream analysis since undefined time-to-event standards exist for these events.

**Clustering.** A heatmap with all the patients and events was generated. We used hierarchical clustering to create dendograms. The distance matrix was generated with the time-to-event matrix, filling all missing events with a negative number (−1000). Distance was calculated with Pearson’s correlation, while linkage was computed as complete.

**Association rules.** The Apriori algorithm [16] was used to discover association rules that are common across multiple patients. Apriori first counts event occurrences to determine the largest 1-item sets. Then, in subsequent steps, it iteratively tries to generate larger candidate item sets, based on an incremental one-step from the previous step, and assign the strength of evidence for that set of events based on the number of occurrences in the database. The Apriori algorithm uses a bottom-up approach. The algorithm finalizes when no candidate list hast larger item sets. The candidate generation step uses a breadth-first search to efficiently count candidate item sets, and prune candidates with infrequent patterns. The algorithm does not consider the time-to-event but only whether the event occurred and in what order it occurred for that patient.

**3. Results**

The epidemiological information obtained pertaining to this one IMSS hospital can be seen in Table 2. The results show the patients with breast cancer in 2014 (N=402), which correlate with national estimates for the country [10]. In this cohort, all patients were female. The highest prevalence of cancer was within the 50-59 year old age group. The majority of patients were diagnosed with early stages of cancer (stages I and II). However, only a few number of patients (N=30) had undergone subtyping with IHC testing in accordance with the Mexican
Consensus. Even in this small cohort, we can still see the same higher prevalence of triple negative breast cancer cases that was previously reported [17]. It is troubling to see that there were 18 (4% of) patients younger than 40 years old.

At IMSS, patients with Breast Imaging Reporting and Data System (BIRADS) reports scoring 1 or 2 are usually deemed low-risk for breast cancer and they have regular annual screenings at their Family Medicine Unit (UMF). Meanwhile, patients with BIRADS reports 0 and 3 are sent to IMSS’s secondary care breast cancer clinic for a closer screening (usually fine-needle biopsy and 6-month screening); while patients with BIRADS reports 4, 5 or 6 are sent directly to the secondary or tertiary care oncology services. At the IMSS-HGR1 hospital that we investigated, 23 patients (6%) had a BIRADS score of 2 or lower, which were not supposed to be sent to the secondary care facility. In these cases, patients had external biopsy investigations in non-IMSS hospitals (usually private), and decided to continue their care for follow-up treatment at the IMSS oncology services.

Table 2. Clinical characteristics of the cohort and receptor status for patients with IHC testing

| Characteristic | Total (%) |
|----------------|-----------|
| Overall N = 402 |           |
| Sex            |           |
| Female         | 402       |
| Male           | 0         |
| Age at diagnosis |     |
| <40            | 18 (4%)   |
| 40-49          | 62 (15%)  |
| 50-59          | 174 (43%) |
| 60-69          | 127 (32%) |
| 70+            | 21 (5%)   |
| BIRADS         |           |
| 0,1,2          | 23 (6%)   |
| 3              | 203 (50%) |
| 4,5,6          | 162 (40%) |
| unknown        | 14 (3%)   |
| Tumor stage    |           |
| I              | 85 (21%)  |
 Patients with IHC testing

|                  | N = 30 |
|------------------|--------|
| **ER testing**   |        |
| Positive         | 17 (57%) |
| Negative         | 11 (37%) |
| Unknown          | 2 (7%) |
| **PR testing**   |        |
| Positive         | 16 (53%) |
| Negative         | 12 (40%) |
| Unknown          | 2 (7%) |
| **HER2 testing** |        |
| Positive         | 5 (17%) |
| Negative         | 23 (77%) |
| Unknown          | 2 (7%) |
| **Additional IHC testing** |        |
| Ki67, P53        | 1 (3%) |
| Unknown          | 29 (97%) |
| **Subtype according to the Consensus** | 14 (47%) |
| Luminal A        | 3 (10%) |
| Luminal B        | 9 (30%) |
| Basal-like / Triple negative | 2 (7%) |
| HER2             | 2 (7%) |

Subtype not enough information to assign

**Clustering analysis**

A hierarchical clustering and heatmap of time-to-events is shown in Figure 2. The time in which each event occurred for each patient is measured using information found in their medical record (in days, positive). To be able to create a dissimilarity matrix to calculate distance between patients, all missing values were assigned a negative value of -1000. It is important to note that this figure does not provide a context for the ordering in which events occurred, but it can still help to provide information about patients with similar trajectories. Three main groups of patients (or clusters) were identified in this graph.
**Figure 2.** Heatmap of time-to-event with patient clustering.

**Patient trajectories**

Medical trajectories for patients in the cohort with IHC testing can be seen in Figure 3, clustered according to the groupings shown in Figure 2. Each patient is represented in a panel (rectangle) with colored bars, indicating the events that a patient experienced in the IMSS hospital. Each row represents one year of follow-up treatment for that patient, which can be from one to four rows (because the time maximum time span is three years and five months). The length of each colored bar represents the time between the occurrence of an event and the event that preceded it. Although events do not occur continuously, but, rather, happen at single time points (e.g. a patient visit, obtaining the results of an x-ray), the visual representation shown in Figure 3 provides a sense of how much time was required for each event to occur, assuming nothing else happened at the same time.
Figure 3. Medical trajectories of the cohort with IHC testing. Each patient is represented by a rectangle, with rows representing each year of follow-up treatment. Events are color-coded according to the type of event, and the length of the bar represents the duration between the occurrence of an event and the event that preceded it.

Turnaround time

The time-to-event for all events is shown in Figure 4. It is measured as the number of days that it took for an event to occur, from the initial visit to the hospital stay (day 0). From this Figure, it is easy to see that visiting the hospital or beginning treatment occurred relatively at the beginning of the patients’ trajectories, while events that typically take place later did in fact occur at a posterior time, such as remission or metastasis. Also, one can see there is a wide distribution of time, when requesting laboratory/imaging testing, which coincides with the continuity of care.
The turnaround time for immunohistochemistry (IHC) testing can be seen in Figure 5. Only 20 patients had information in their medical records relating to the timing of IHC testing. Although this service is referred to in the IMSS medical records, it is subcontracted to a private laboratory. There was large variation surrounding two key variables: a) the time needed to request IHC testing relative to the first day of follow-up treatment at the hospital, and b) the turnaround time to obtain those results. For the first one, the average time of request was 117 days (95% C.I. 80 – 154 days). For the second one, the average turnaround time for obtaining those results was 56 days (95% C.I. 36 – 77 days).

There were 15 out of 20 patients whose treatments were adjusted, in accordance with IMSS protocol, after obtaining the results of the IHC testing. Overall, for the 30 patients with
IHC testing, 17 patients (56.7%) had their treatments adjusted, and 13 patients (43.3%) had their treatments confirmed.

**Figure 5.** Turnaround times for IHC testing. The size of the bar indicates time in days from requesting results for IHC testing to obtaining them. For each patient, the initial time is the first day of follow-up treatment at the hospital. The color of the bar indicates the quartile within which the data falls in the distribution. The diamond mark indicates that the treatment was adjusted, according to protocols, after obtaining IHC results. The blue dashed line indicates the 1st quartile for the time it took before an IHC request was submitted. Violin plots with whiskers are correspondingly provided for the time to IHC request (from start to request), time to IHC results (from start to the obtaining of results), and turnaround time of IHC testing (from request to results).

**Association rules**

Association rules were mined from the events of this cohort using the Apriori algorithm. The five top-ranking association rules are shown in Figure 6. These rules have a support of
63.3% of cases, with a confidence of 90.5% and a rule performance (lift, or precision) of 1.3. For rules with lift larger than 1, it can be assumed that the right-hand side of the rule is a consequence of the left-hand side. In our mined rules, we can observe that, in general, IHC testing, particularly ER/PR testing, results in adjusting treatment plans.

**Figure 6.** Top-5 association rules mined with the Apriori algorithm

### 4. Discussion

#### Key findings

Breast cancer in Mexico (and worldwide) continues to be a public health concern, and increasing research is being done on the characterization of better genomic variants. Although our study investigated a small number of patients, the prevalence and epidemiological characteristics among our cohort are similar to what has previously been estimated for the country (i.e. in regards to age group and staging) [10]. Our study further investigates the timeliness of IHC testing, so our conclusions can then be used to inform hospital administrators and public health officials around the world about the importance of timely IHC testing.
On the therapeutic value. The therapeutic importance of immunophenotyping (the use of IHC testing for subtyping of cancer patients) needs to be carefully considered and addressed in treatment plans by oncologists. IMSS protocol states that estrogen positive (ER+) and/or progesterone positive (PR+) patients could benefit from hormone replacement therapy to prevent recurrence. For the patient, that usually means that the traditional FEC chemotherapy regimen composed by 5-fluorouracil, epirubicin, and cyclophosphamide, would be adjusted to include some hormone replacement therapy, either aromatase inhibitors to block estrogen production (e.g. anastrozole, letrozole, or exemestane), or some drug that could interfere with the ability of estrogen to stimulate the growth of breast cells (e.g. tamoxifene or toremifene).

In contrast, patients with HER2 positive (HER2+), which tumors tend to grow and spread more aggressively, could benefit from a targeted therapy that could prescribe drugs to block HER2 (e.g. trastuzumab or pertuzumab). However, the use of these drugs can have different side effects than traditional chemotherapy. The use of in situ hybridization (ISH), instead of IHC, can be used to determine HER2 status with an overall concordance with IHC, and it may be more beneficial to use both [18]. There is a need to balance sensitivity of the study with hospital workload and expenses. Furthermore, HER2 status has been successfully incorporated into medical practice to guide treatment decisions for breast cancer patients [19]. In fact, the American Society of Clinical Oncology/College of American Pathologists (ASCO/CAP) updated their 2013 guidelines to designate more patients as eligible for trastuzumab therapy, in accordance with ISH and IHC testing [20].

On the timeliness of testing. Opportune IHC testing, understood as the availability of IHC results before the beginning of any treatment, is of critical importance. For IMSS, the timeliness of IHC results were typically not opportune, but whenever they were available, they triggered a
response from the clinical oncology team to adjust patient treatments. The widespread
distribution of time to request and turnaround time demonstrates the lack of standardization of
this process. Many factors could have contributed to the delay in IHC testing, including: patient
non-adherence to appointments; hospital overflow, resulting in longer appointments; logistical
problems relating to IHC testing’s status an external service to the IMSS hospital of this study;
and administrative problems in the hospital resulting in inadequate tracking of tests and results.

On the need for improved clinical guidelines. It is important to establish an appropriate
timeline for opportune IHC testing. For example, In the United States, the College of American
Pathologists’ (CAP) guideline on turnaround time for standard biopsies is around two days for
90% of cases [21]. In fact, the joint guideline on HER2 testing from the American Society of
Clinical Oncology (ASCO) and CAP recommends informing the patients about the expected
turnaround time [22]. In Australia, a study with more than 78,000 patients tested for HER2
status, with both ISH and IHC, revealed that the average turnaround time was between 4-5 days,
which decreased due to the increased volume of patients being tested [23]. In Saudi Arabia, a
study in 2015 showed that 24% of cases fall outside the recommended CAP turnaround time
[24]. The European Society for Medical Oncology (ESMO) published a survey on 24 European
countries were the turnaround time was 10 days or less for 89% of laboratories [25]. In the
United Kingdom, the Royal College of Pathologists recommend the use of key performance
indicators to have a histopathology diagnosis within seven days of biopsy in 90% of cases [26].

Given their resource-available settings, clinical guidelines in developed countries do not
have a recommendation regarding the maximum turnaround time that could be effective in a
patient’s treatment trajectory. More importantly, they do not address the time between initial
diagnosis, molecular testing (either via IHC or genetics), and the selection of treatment. In
Mexico, clinical guidelines for breast cancer recommend testing for ER, PR, and HER2 as part of the histopathology study, but fail to provide guidance regarding turnaround time [13]. IMSS maintains a medical procedures manual, which includes ten key indicators for breast cancer screening, diagnosis, and treatment [27]. This manual measures time-to-diagnosis in a 30-day period, including imaging through mammography and results of histopathology report. In addition, the institution also measures time-to-treatment measured from the date of diagnosis, which should be achieved within a 21-day period. Recognizing the resource constraints of IMSS, the implementation of a key indicator policy related to IHC testing might significantly reduce turnaround times.

Limitations

On the need for accurate electronic health records (EHR). Missing information was common across the paper and electronic records in our study. Clinicians and administrative staff at the IMSS hospital are still getting used to the novel implementation of the EHR, which had an impact on our ability to better characterize this cohort. In the absence of a cancer registry, our best estimate of the disease is the institutional census. As the hospitals in Mexico, and elsewhere, continue to become more and more electronic, there is a need to develop better software tools to analyze the information obtained, including medical natural language processing and machine learning applications.

On the visual representation. The medical trajectories shown in Figure 1 are useful in quickly providing a patient history overview of hospital care. With more work on the user interface, we envision this tool could eventually represent a valuable visual aid in which a patient and their companions might be able to better communicate with their clinicians about the
management of their disease (it could probably also appear in a printed version). Currently, some of the challenges of this tool include the missing identification of events that are overlapping, and the impossibility of further elaborating on the details of each event. For a hospital administrator, Figure 1 can provide a quick overview of the patients seen in their hospital, which could be used as a decision-making tool, with the proper validation. This visual aid creation tool can be obtained from the repository at https://github.com/arturolp/patientTrajectory.git

**Healthcare system implications**

Adverse events in the trajectory of an oncological patient, which might include hospitalization related to cancer, recurrence of tumor, or metastasis, are extremely costly for the healthcare system. The use of IHC testing was shown in our study to help with the selection of precise treatment for patients (either by adjusting the treatment or confirming it). The association rules found in our study confirm that ER and PR testing are associated with the adjustment of treatment. However, the time in which IHC testing is performed is of critical importance if we want to influence improved prognosis.

IHC testing is being carried out as an external service at the IMSS hospital in our study. As the Mexican healthcare system continues to transition from reactive to preventative care, the need for more IHC testing in breast cancer and other diseases will certainly allow for the further development of its own testing facilities, therefore allowing for some economy of scale.

We have shown that the use of surrogate immunohistochemistry, as described by the Mexican Consensus and previous literature worldwide, can have a beneficial therapeutic effect on breast cancer patients. The aims of any healthcare system should be the identification of earlier events that can have an impact on downstream events in the trajectory of an oncological
patient’s treatment. In resource-constrained settings, it is important not only to consider surrogate alternatives to more costly diagnostics (e.g. genomic testing), but also to incorporate the regulatory and logistical aspects of implementing these surrogate IHC tests. IMSS must face the important challenge of continuing to improve their turnaround times, which should have a positive impact on the prognosis of their patients. Finally, other healthcare systems would also benefit from determining the maximum turnaround time in which IHC testing can still be opportune and have a therapeutic beneficial effect.

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Authors’ contributions

Conceptualization: MFRM, ALP, and CAA
Data curation: MFRM
Formal analysis: MFRM and ALP
Funding acquisition: CDB
Investigation: MRFM, ALP, SMFV, and CAA
Methodology: ALP
Resources: RAR and CDB
Software: ALP
Supervision: CDB, RAR, and CAA
Visualization: MFRM, ALP
Writing – original draft: ALP
Writing – review & editing: MFRM, CAA, SMFV, RAR, CDB, ALP

Data Availability

Data belongs to the Mexican Institute of Social Security (IMSS), through its state Delegation of Michoacan. IMSS’s Coordination of Health Research may grant access to this data on a case-by-case basis to researchers who obtain the necessary approvals from IMSS’s National Scientific Research Committee and National Bioethics Committee.

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Competing interests

RAR declares to be IMSS’s state delegate in Michoacan, overseeing the hospital mentioned in this study. The remaining authors declare no conflicts of interest.

References

1. Sørlie T, Perou CM, Tibshirani R, Aas T, Geisler S, Johnsen H, et al. Gene expression patterns of breast carcinomas distinguish tumor subclasses with clinical implications. Proc Natl Acad Sci U S A. National Acad Sciences; 2001;98: 10869–10874. doi:10.1073/pnas.191367098

2. Abu-Khalf M, Pusztai L. Influence of genomics on adjuvant treatments for pre-invasive and invasive breast cancer. Breast. 2013;22 Suppl 2: S83–7. doi:10.1016/j.breast.2013.07.015

3. Enewold L, Geiger AM, Zujewski J, Harlan LC. Oncotype Dx assay and breast cancer in the United States: usage and concordance with chemotherapy. Breast Cancer Res Treat. Springer US; 2015;151: 149–156. doi:10.1007/s10549-015-3366-7

4. Lips EH, Mulder L, de Ronde JJ, Mandjes IAM, Koolen BB, Wessels LFA, et al. Breast cancer subtyping by immunohistochemistry and histological grade outperforms breast cancer intrinsic subtypes in predicting neoadjuvant chemotherapy response. Breast Cancer Res Treat. Springer US; 2013;140: 63–71. doi:10.1007/s10549-013-2620-0

5. Won JR, Gao D, Chow C, Cheng J, Lau SYH, Ellis MJ, et al. A survey of immunohistochemical biomarkers for basal-like breast cancer against a gene expression
profile gold standard. Mod Pathol. 2013;26: 1438–1450. doi:10.1038/modpathol.2013.97

6. Cardenas Sanchez J, Bargallo Rocha JE, Erazo Valle A, Poitevin Chacon A, Valero Castillo V, Perez-Sanchez V. [Consenso Mexicano sobre diagnóstico y tratamiento del cáncer mamario]. 6 ed. 2015. pp. 1–149.

7. Lara-Medina F, Perez-Sanchez V, Saavedra-Perez D, Blake-Cerda M, Arce C, Motola-Kuba D, et al. Triple-negative breast cancer in Hispanic patients: high prevalence, poor prognosis, and association with menopausal status, body mass index, and parity. Cancer. 2011;117: 3658–3669. doi:10.1002/cncr.25961

8. Robles-Castillo J, Ruvalcaba-Limón E. [Cáncer de mama en mujeres mexicanas menores de 40 años]. Ginecologia y Obstetricia de Mexico. 2011;79: 482–488.

9. Villarreal-Garza C, Alvarez-Gomez RM, Perez-Plasencia C, Herrera LA, Herzog J, Castillo D, et al. Significant clinical impact of recurrent BRCA1 and BRCA2 mutations in Mexico. Cancer. 2015;121: 372–378. doi:10.1002/cncr.29058

10. Mohar-Betancourt A, Reynoso-Noveron N, Armas-Texta D, Gutierrez-Delgado C, Torres-Dominguez JA. Cancer Trends in Mexico: Essential Data for the Creation and Follow-Up of Public Policies. Journal of Global Oncology. 2017;: JGO.2016.007476–9. doi:10.1200/JGO.2016.007476

11. Secretaria de Salud de Mexico. [Diagnóstico y Tratamiento de la Patología Mamaria Benigna en Primer y Segundo Nivel de Atención]. 2009;: 1–66.

12. Secretaria de Salud de Mexico. [Prevention, Screening, and Timely Referral of Suspected Breast Cancer in the First Level of Care]. 2011 pp. 1–74.

13. Secretaria de Salud de Mexico. [Diagnóstico y Tratamiento del Cáncer de Mama en Segundo y Tercer Nivel de Atención]. 2012;: 1–102.
14. Instituto Nacional de Estadística y Geografía. [Encuesta Intercensal 2015]. 2015.

15. Parkes J, Hendricks M, Ssenyonga P, Mugamba J, Molyneux E, Schouten van Meeteren A, et al. SIOP PODC adapted treatment recommendations for standard-risk medulloblastoma in low and middle income settings. Pediatric Blood &amp; Cancer. 2015;62: 553–564. doi:10.1002/pbc.25313

16. Agrawal R, Srikant R. Fast algorithms for mining association rules. Proc 20th int conf very large data bases. 1994.

17. Villarreal-Garza C, Weitzel JN, Llacuachaqui M, Sifuentes E, Magallanes-Hoyos MC, Gallardo L, et al. The prevalence of BRCA1 and BRCA2 mutations among young Mexican women with triple-negative breast cancer. Breast Cancer Res Treat. Springer US; 2015;150: 389–394. doi:10.1007/s10549-015-3312-8

18. Solomon JP, Dell'Aquila M, Fadare O, Hasteh F. Her2/neu Status Determination in Breast Cancer: A Single Institutional Experience Using a Dual-Testing Approach With Immunohistochemistry and Fluorescence In Situ Hybridization. Am J Clin Pathol. 2017;147: 432–437. doi:10.1093/ajcp/aqw224

19. Goddard KAB, Bowles EJA, Feigelson HS, Habel LA, Alford SH, McCarty CA, et al. Utilization of HER2 genetic testing in a multi-institutional observational study. Am J Manag Care. NIH Public Access; 2012;18: 704–712.

20. Lim TH, Lim AST, Thike AA, Tien SL, Tan PH. Implications of the Updated 2013 American Society of Clinical Oncology/College of American Pathologists Guideline Recommendations on Human Epidermal Growth Factor Receptor 2 Gene Testing Using Immunohistochemistry and Fluorescence In Situ Hybridization for Breast Cancer. Arch Pathol Lab Med. 2016;140: 140–147. doi:10.5858/arpa.2015-0108-OA
21. Novis DA, Zarbo RJ, Saladino AJ. Interinstitutional comparison of surgical biopsy diagnosis turnaround time: a College of American Pathologists Q-Probes study of 5384 surgical biopsies in 157 small hospitals. Arch Pathol Lab Med. 1998;122: 951–956.

22. Wolff AC, Hammond MEH, Hicks DG, Dowsett M, McShane LM, Allison KH, et al. Recommendations for Human Epidermal Growth Factor Receptor 2 Testing in Breast Cancer: American Society of Clinical Oncology/College of American Pathologists Clinical Practice Guideline Update. J Clin Oncol. 2013;31: 3997–4013. doi:10.1200/JCO.2013.50.9984

23. Morey AL, Brown B, Farshid G, Fox SB, Francis GD, McCue G, et al. Determining HER2 (ERBB2) amplification status in women with breast cancer: final results from the Australian in situ hybridisation program. Pathology. Elsevier; 2016;48: 535–542. doi:10.1016/j.pathol.2016.05.007

24. Alshieban S, Al-Surimi K. Reducing turnaround time of surgical pathology reports in pathology and laboratory medicine departments. BMJ Qual Improv Rep. BMJ Open Quality; 2015;4: u209223.w3773. doi:10.1136/bmjquality.u209223.w3773

25. Boleij A, Tembuyser L, Taylor A, Kafatos G, Jenkins-Anderson S, Tack V, et al. PD-015A survey on current RAS-mutation testing practices in Europe. Ann Oncol. Oxford University Press; 2015;26: iv105–iv105. doi:10.1093/annonc/mdv234.14

26. Pathologists TRCO. Key Performance Indicators in Pathology. 2013;; 1–23.

27. Instituto Mexicano del Seguro Social. [Manual Metodológico de Indicadores Médicos 2016]. 2016;; 1–452.
Cumulative patients with confirmed breast cancer with visits to HGR1 in 2014 (Institutional census)

N = 402

Without IHC testing (Jan 2014 – Mar 2017)
N = 372

Deceased (Jan 2014 – Mar 2017)
N = 0

With IHC testing (Jan 2014 – Mar 2017)
N = 30
