Human & Veterinary Medicine
International Journal of the Bioflux Society HVM

OPEN ACCESS
Research Article

A screening for the candidate diseases that could benefit from immunotherapy

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Abstract. Objective: Genomic instability is one of the hallmarks of cancer and has seen an increased interest in the last decades due to its capability to predict the response to immunotherapy. The aim of the current study was to determine the microsatellite instability (MSI) on a local cohort and, on the diseases that present Microsatellite Instability High (MSI-H), to further evaluate the efficacy of these drugs on a publicly available cohort. Material and Method: This was a cross-sectional, observational and analytical study in which we included the patients from our institution and also from the TMB and Immunotherapy online cohort. Results: In the current study we observed that the most relevant cases for which we could implement immunotherapy in the treatment of MSI high patients are represented by the colon cancer cases. This is supported by the fact that colon cancer patients represent the highest percentage of malignancies we treat in our unit, which have a clinically relevant prevalence of MSI high. Conclusion: Thus, in the current study we observed that immunotherapy might be a useful tool in the treatment of metastatic colon cancer in our unit.

Key Words: genomic instability, colon cancer, immunotherapy

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Introduction

One of the hallmarks of cancer is represented by genomic instability. This is associated with a hypermutation status that is able to increase the probability for driver mutations to appear. Nonetheless, this hypermutation status is also associated with the production of neoantigens, which can be detected by the immune system, representing one of the main concepts in tumor immunology (Hanahan & Weinberg 2000, 2011). This was the basis of the therapy and from the early 2000s, checkpoint inhibitors have been implemented in clinical trials with success, starting with melanoma patients. (Leonardi et al2020). Because of the important impact that this trial has had on melanoma, other trials have also been started, with several of them showing checkpoint inhibitors having a high efficiency and a low incidence of severe side-effects (Gaynor et al 2020; Sharon et al 2014). This has culminated with the approval of using Pembrolizumab for solid tumors in which genomic instability could be observed (Marcus et al 2019). This represents an important step in the direction of personalized medicine, as treatments are based more frequently on the molecular profile of a disease, leading to better results. However, these results are based on large clinical trials and other studies will still have to be performed in order to determine the diseases in which checkpoint inhibitors are useful at a local level. Furthermore, although these drugs have been approved in some countries, they are not approved universally, so certain procedures still have to be undertaken. Thus, it is important to determine in which of the diseases would these drugs be most efficient at a local level.

The aim of the current study was to determine the microsatellite instability (MSI) on a local cohort and, on the diseases that present MSI-high, to further evaluate the efficacy of these drugs on a publicly available cohort.

Material and Methods

This was a cross-sectional, observational and analytical study. In the present study we included patients with digestive or gynecological malignancies, who were admitted at the Alba Iulia County Emergency Hospital in the time period august 2018-september 2020.

This study was conducted in accordance with the Declaration of Helsinki and with the approval of the Ethics Committee from the University of Medicine and Pharmacy "Iuliu Hatieganu" Cluj Napoca (nr.303/26.07.2018).

MSI testing was performed by Resident Laboratory Oradea. A number of 5 microsatellites were used (BAT-25, BAT-26, NR-21, NR-24, MONO-27), and the tests were performed from both tumoral tissue and normal control tissue. If none of the five markers showed instability, the tumor was referred to as MS-stable (MSS); if only one marker showed instability, then the tumor was defined as MSI-low (MSI-L); and if instability was revealed in two or more markers, the tumor was defined as MSI-high (MSI-H).

TMB and Immunotherapy (MSKCC Nat Genet 2019) (Samstein et al 2019) data was downloaded using the cBioPortal interface (Cerami et al 2012; Gao et al 2013). We dichotomized tumor

Table 1. Differences between patients considering the TMB. COAD = colon cancer; STAD = stomach cancer

Stage and Disease Group		Primary COAD n=37			Metastatic COAD n=48			Primary STAD n=22			Metastatic STAD n=13		
TMB Group		High n=14	Low n=23	p	High n=14	Low n=34	p	High n=4	Low n=18	p	High n=1	Low n=12	p
Malegender		8(57.1%)	9(39.1%)	0.328	10(71.4%)	22(64.7%)	0.746	3(75.0%)	14(77.8%)	1	0(0%)	8(66.7%)	0.385
Age(years)		61(54.65)	53(45.59)	0.097	62(59.68)	53(46.61)	0.029	71(68.76)	62(48.70)	0.067	71(71.71)	50(40.70)	0.422
Metastasissite	Lymph node	NA	NA	NA	4(28.6%)	3(8.8%)		NA	NA	NA	0(0%)	1(8.3%)	
	Liver	NA	NA	NA	3(21.4%)	20(58.8%)	0.038	NA	NA	NA	0(0%)	0(0%)	1
	Other	NA	NA	NA	7(50.0%)	11(32.4%)		NA	NA	NA	1(100%)	11(91.7%)	

mutational burden (TMB) using the cutoff of 20, with samples over 20 being considered to have a high genomic instability. Data analysis was performed using R 4.0.1. Categorical variables were represented as absolute value (percent). Contingency tables were analyzed using Fisher's test. Considering the low number of patients in the final groups, we decided to use non-parametric statistics. Continuous variables were represented as median (quartile 1, quartile 3). Differences between two non-normally distributed groups were performed using Mann-Whitney-Wilcoxon rank sum test. Kaplan Meyer curves were used to represent survival. Log-rank test was used to assess survival. A p-value under 0.05 was considered statistically significant.

Results

From our hospital we included a total of 52 patients, of which 20 (38.5%) had colon cancer, 4 (7.7%) had stomach cancer, 2 (3.8%) had pancreatic cancer, 9 (17.3%) had ovarian cancer, 8 (15.4%) had breast cancer, 6 (11.5%) had endometrium cancer, 2 (3.8%) had vulva cancer and 1 (1.9%) had cervix cancer. Of these patients, 1 (50%) patient with endometrium cancer had MSI low, 2 (50%) patients with stomach cancer had MSI high and 3 (15%) patients with colon cancer had MSI high. Because of this, we decided to further investigate the implication of genomic instability expressed through TMB in colon and stomach cancers regarding survival and metastasis status on the TMB and Immunotherapy (MSKCC Nat Genet 2019) cohort. There was no difference between primary and metastatic samples considering the TMB in the case of colon cancer (p = 0.487), nor in the case of stomach cancer (p = 0.630). We described the clinical differences between patients in Table 1.

In the survival analysis there was no statistical difference when considering primary (p = 0.660), or the metastatic samples (p = 0.550) divided by TMB. Nonetheless, we did observe a tendency for statistical significance in the survival analysis of colon primary samples divided by TMB (Figure 1) and we observed there was a statistically significant difference regarding the survival analysis of colon cancer metastatic samples divided by TMB (Figure 2).

Discussions

In the current study we observed that the most relevant cases for which we could implement immunotherapy in the treatment of MSI high patients are represented by the colon cancer cases. This is supported by the fact that colon cancer patients represent the highest percentage of malignancies treated in our

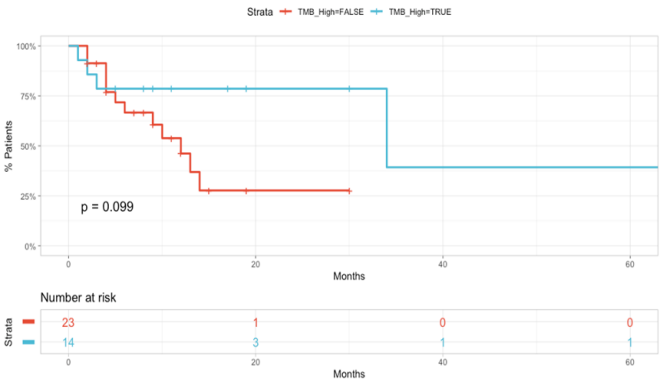

Fig. 1. Overall survival of patients with colon cancer divided by the TMB analyzed from the primary samples

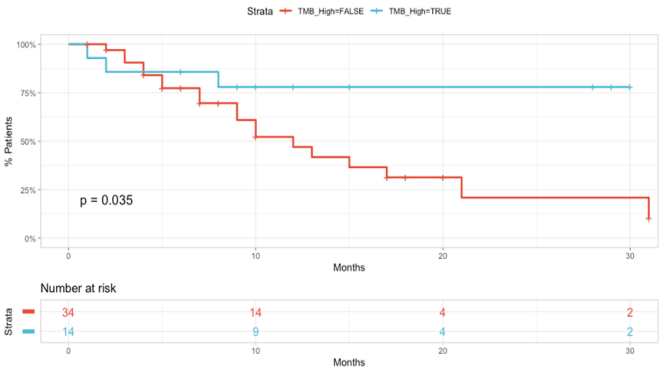

Fig. 2. Overall survival of patients with colon cancer divided by the TMB analyzed from the metastatic samples

unit, which have a clinically relevant prevalence of MSI high. More than this, on the online cohort we observed in the case of metastatic colon cancer that high TMB is associated with a better prognosis when treated with immunotherapy, whereas, in the case of primary colon cancer samples, the survival analysis only had a tendency for statistical significance considering the TMB dichotomization, with the trend being maintained as in the case of primary samples. We also observed that metastatic colon cancer patients with high TMB had a higher age, a higher frequency of lymph node metastases and a lower frequency of liver metastases, this showing additional differences secondary to the biology that is influenced by genomic instability.

Several clinical trials were conducted using immunotherapy in the treatment of colon cancer. There were trials that yielded encouraging results with immunotherapy used in the neoadjuvant setting and being able to induce a response in all the included

cases (Chalabi et al 2020). Conversely, there were also trials in which Pembrolizumab induced a partial response in only one patient with PD-L1 expression and MSI high (O'Neil et al 2017). This type of trial got expanded so that it could be suitable for the treatment of metastatic colon cancer (Le et al 2017; Overman et al 2017). In addition to this, although MSI high was shown to be associated with response to immunotherapy (Le et al 2017) other trials have also been started which included colon cancer with microsatellite stability (NCT03642067, NCT03647839, NCT03007407, NCT03396926, NCT03631407, NCT03608046, NCT03800602).

The strength of the current manuscript consists in the arising interest for the further development of this therapeutic approach for patients with colon cancer.

The limitation of the study is represented by the low number of patients included and the heterogeneous nature of the selected diseases.

Conclusion

In the current study we observed that immunotherapy might be a useful tool in the treatment of metastatic colon cancer in our unit.

References

Cerami E, Gao J, Dogrusoz U, Gross B, Sumer S, et al. The cBio Cancer Genomics Portal: An Open Platform for Exploring Multidimensional Cancer Genomics Data: Figure 1. Cancer Discovery 2012; 2(5), 401–404. doi: 10.1158/2159

Chalabi M, Fanchi LF, Dijkstra KK, et al. Neoadjuvant immunotherapy leads to pathological responses in MMR-proficient and MMR-deficient early-stage colon cancers. Nat Med 2020; 26, 566–576. https://doi.org/10.1038/s41591-020-0805-8

Gao J, Aksoy BA, Dogrusoz U, Dresdner G, Gross B, et al. Integrative Analysis of Complex Cancer Genomics and Clinical Profiles Using the cBioPortal. Science Signaling 2013; 6(269), pp. pl1. doi: 10.1126/scisignal.2004088

Gaynor N, Crown J, Collins DM, et al. Immune checkpoint inhibitors: Key trials and an emerging role in breast cancer. Seminars in Cancer Biology 2020; https://doi.org/10.1016/j.semcancer.2020.06.016

Hanahan D, Weinberg RA, et al. The Hallmarks of Cancer. Cell, 100(1), 2000; 57–70. https://doi.org/10.1016/S0092-8674(00)81683-9

Hanahan D, Weinberg RA, et al. Hallmarks of Cancer: The Next Generation. Cell, 2011; 144(5), 646–674. https://doi.org/10.1016/j.cell.2011.02.013

Le DT, Durham JN, Smith KN, Wang H, Bartlett BR, Aulakh LK, et al. Mismatch repair deficiency predicts response of solid tumors to PD-1 blockade. Science 2017; 357(6349), 409–413. doi: 10.1126/science.aan 6733

Leonardi G, Candido S, Falzone L, Spandidos D, et al. Cutaneous melanoma and the immunotherapy revolution (Review). International Journal of Oncology 2020; 57(3), 609–618. https://doi.org/10.3892/ijo.2020.5088

Marcus L, Lemery SJ, Keegan P, et al. FDA Approval Summary: Pembrolizumab for the Treatment of Microsatellite Instability-High Solid Tumors. Clinical Cancer Research 2019; 25(13), 3753–3758. doi: 10.1158/1078-0432.CCR-18-4070

O'Neil BH, Wallmark JM, Lorente D, Elez E, Raimbourg J, Gomez-Roca C, et al. Safety and antitumor activity of the anti–PD-1 antibody pembrolizumab in patients with advanced colorectal carcinoma. PLOS ONE 2017; 12(12). https://doi.org/10.1371/journal.pone.0189848

Overman MJ, McDermott R, Leach JL, Lonardi S, Lenz HJ, et al. Nivolumab in patients with metastatic DNA mismatch repair-deficient or microsatellite instability-high colorectal cancer (CheckMate 142): An open-label, multicentre, phase 2 study. The Lancet Oncology 2017; 18(9), 1182–1191. https://doi.org/10.1016/S1470-2045(17)30422-9

Samstein RM, Lee CH, Shoushtari AN, Hellmann MD, Shen R, et al. Tumor mutational load predicts survival after immunotherapy across multiple cancer types. Nature Genetics 2019; 51(2), 202–206. https://doi.org/10.1038/s41588-018-0312-8

Sharon E, Streicher H, Goncalves P, et al. Immune checkpoints in cancer clinical trials. Chinese Journal of Cancer 2014; 33(9), 434–444. doi:10.5732/cjc.014.10122

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Citation	Pitu F, Pasca S, Curca R, Al Hajjar N. A screening for the candidate diseases that could benefit from immunotherapy. HVM Bioflux 2021;13(2):67-69.
Editor	Antonia Macarie
Received	24 January 2021
Accepted	2 March 2021
Published Online	19 April 2021
Funding	None reported
Conflicts/ Competing Interests	None reported