Screening and Management of Lynch Syndrome: The Chinese Experience

Jiehai Yu, MD[1] Pei-Rong Ding, MD, PhD[1] Wu Jiang, MD[1]

[1] Department of Colorectal Surgery, State Key Laboratory of Oncology in South China, Collaborative Innovation Center for Cancer Medicine, Sun Yat-sen University Cancer Center, Guangzhou Guangdong, P. R. China

Clin Colon Rectal Surg 2023;36:369–377.

Address for correspondence Wu Jiang, MD, Department of Colorectal Surgery, State Key Laboratory of Oncology in South China, Collaborative Innovation Center for Cancer Medicine, Sun Yat-sen University Cancer Center, 651 Dongfeng East Road, Guangzhou 510060, P. R. China (e-mail: Jiangwu@sysucc.org.cn).

Abstract

Lynch syndrome (LS), caused by germline mutations in the mismatch repair genes, is the most common hereditary colorectal cancer. While LS is also associated with various cancers, early detection of the proband is meaningful for tumor prevention, treatment, and familial management. It has been a dramatic shift on the screening approaches for LS. As the rapid development of the molecular biological methods, a comprehensive understanding of the LS screening strategies will help to improve the clinical care for this systematic disease. The current screening strategies have been well validated but mainly by evidence derived from western population, lacking consideration of the ethnic heterogeneity, which hampers the universality and clinical application in China. Hence, this review will focus on the Chinese experience in LS screening, aiming to help better understand the ethnic diversity and further optimize the screening strategies.

Keywords
- colorectal cancer
- Lynch syndrome
- universal screening
- ethnic heterogeneity

Hereditary colorectal cancer (CRC) accounts for 7 to 10% of all the CRC.[1,2] Among these familial CRC cases, Lynch syndrome (LS), caused by the germline alterations of the mismatch repairing (MMR) genes (MLH1, MSH2, MSH6, and PMS2), or deletions in the 5′ area of EPCAM is the most common one, and it accounts for 2 to 4% of all the CRC.[1,3–6] Apart from CRC, patients with LS are susceptible to multiple gynecological cancers such as endometrial cancer, ovary cancer, and other gastrointestinal cancers like gastric cancer and pancreatic cancer at a young age.[7–9] Therefore, early identification of the LS proband will greatly contribute to better prevention, treatment, and familial management of this systematic disease and this relies on effective screening. In the past decades, different screening strategies including guideline based on clinical criteria and universal screening, have been implemented to improve the efficiency of LS individual detection.[9–12]

Owing to the rapid development of molecular testing technique, the sensitivity and specificity of LS individual identification have increased and the screening strategies become more optimized. However, because of the increasingly common use of biological molecular methods, the regional and racial heterogeneities of LS have come into the spotlight. The genetic mutation characteristics and the subsequently screening strategies derived from the occidental are not fit in the Chinese population perfectly.

Herein, the LS screening strategy and management development in China is reviewed here, aiming to help better understand the heterogeneity of LS and therefore provide more comprehensive evidence to optimize the screening strategy of LS.

Epidemiology, Clinical, and Pathological Features of LS

Among the four MMR genes, germline alterations in MLH1 and MSH2 account for about three-quarters of cases, while mutations in MSH6 and PMS2 are relatively rare.[1,4,13] On the other hand, in the overall population, interestingly, the most common pathogenic germline variant was found to be PMS2

article published online May 3, 2023

Issue Theme Application of Molecular Profiling in Colorectal Cancer Surgery; Guest Editor: Pei-Rong Ding, MD, PhD

DOI https://doi.org/10.1055/s-0043-1767706.
ISSN 1531-0043.

Table 1 Studies on the prevalence of Lynch syndrome

Reference/Nationality	No. of cases	Study setting	Prevalence of LS
Salovaara et al[18] (Finland)	1,044	Population	2.7%
Hampel et al[19] (USA)	1,566	Population	2.8%
Yurgelun et al[1] (USA)	1,058	Population	3.1%
Pérez-Carbonell et al[20] (Spain)	2,093	Population	0.7%
Chika et al[44] (Japan)	1,234	Single-institution	0.9%
Jiang et al[21] (China)	3,139	Single-institution	2.9%
Yang et al[22] (China)	4,966	Multi-institutions	1.9%

Abbreviation: LS, Lynch syndrome.

(0.140%), followed by MSH6 (0.132%), MLH1 (0.051%), and MSH2 (0.035%) in a multireligion epidemiologic study investigating 5,744 CRC patients and 37,634 first-degree relatives in the United States, Canada, and Australia. And the frequency of any MMR gene alteration was 0.359%.[14] This discordance may result from the minor risk of cancer for PMS2 and MSH6 comparing to the other two MMR genes.[8,15–17] The prevalence of LS CRC ranges from 0.7 to 2.8% in western countries[18–20] and it is reported to be approximately 1.9 to 2.9% in China[21,22] (►**Table 1**). And the mutation proportion of the responsible genes in Chinese population is similar to that of the west, in other words, variants in MLH1 and MSH2 occupy the majority.[21]

The clinical and pathological features of LS-associated CRC are distinct from those of sporadic CRC. First, the onset age of LS CRC is relatively earlier than that of sporadic CRC,[23] with a mean onset age of around 43 years.[24] Second, LS-associated CRC tends to be located in the right-sided/proximal colon. Third, the prevalence of multiple primary CRC in LS patients is also higher and meanwhile they also have increasing risk of developing another CRC after the primary CRC. In view of this, more frequent colonoscopy surveillance is recommended for LS carriers[25,26] and also more extensive surgery should be taken into consideration for patients with a screening-detected primary CRC and with a family history of LS.[27] Additionally, the prognosis of LS CRC patients is reported to be more favorable than that of sporadic CRC patients with the same stage based on several retrospective studies,[28] whereas this conclusion requires further verification due to the potential selection bias of those studies. In terms of histopathology, LS-associated CRC commonly presents with poor-differentiated, mucinous/signet-ring, or medullary adenocarcinoma, together with higher frequency of Crohn's-like reaction and more tumor-infiltrating lymphocytes.[29,30]

Development of LS Screening in China

The screening evaluation for LS carriers goes through two major stages and nowadays has dramatically shifted from clinical criteria to universal screening (►**Fig. 1**), benefitting from the application of molecular biological methods and particularly the progression in the next-generation sequencing (NGS).

Clinical Criteria

The clinical criteria-based diagnostic strategy is mainly based on clinical features including age of onset, number of tumors, and family history. The representative criteria are the Amsterdam criteria I, described in 1991 by Vasen et al[31] and were revised as Amsterdam criteria II in 1999.[10] However, since these criteria are highly dependent on the family history and the standards are relatively rigid, up to 68% individuals missed diagnosis.[32] And this become worse in China where the family size is relatively small and as such the family history tends to be vague, which indicates up to 90% of LS individuals cannot be detected using the Amsterdam criteria in China.[21] As early as in 1996, Yuan et al found germline mutations in 7 families out of 31 families which did not fulfill the Amsterdam criteria but were highly suspected of LS due to their early onset ages in China,[33] revealing the lack of global universality of those criteria. Then, the suspected hereditary nonpolyposis CRC criteria (sHNPCC) I were issued by them to facilitate the detection for small families and the detailed items included: (1) vertical transmission of CRC or at least two siblings affected by CRC in a family; and (2) development of multiple colorectal tumors, or at least one CRC diagnosed before the age of 50 years, or development of extracolonic cancer (such as endometrium, stomach, small intestine, ovary, hepatobiliary, and urinary tract cancer) in family members. Subsequently, the clinical and genetic manifestations were found similar by comparing 29 international collaborative group-HNPCC (ICG-HNPCC) families fulfilling the Amsterdam criteria and 34 sHNPCC families fulfilling the sHNPCC criteria. Notably, the ICG group had more CRC patients per family than the suspected group (4.07 vs. 2.44, $p < 0.05$), indicating sHNPCC criteria may facilitate the detection of LS individuals in small families[34] and later sHNPCC criteria became the Chinese criteria for LS (►**Table 2**).

Screening strategy based only on the clinical features exhibit a low sensitivity and may not be universal worldwide. Hence, the United States National Cancer Institute proposed the Bethesda criteria and the modified version in 1996 and 2002, respectively.[11,12] Comparing to the Amsterdam criteria, the Bethesda guidelines have relatively relaxing standards for family history and age of onset. Instead, it takes pathological characteristics into consideration, and further

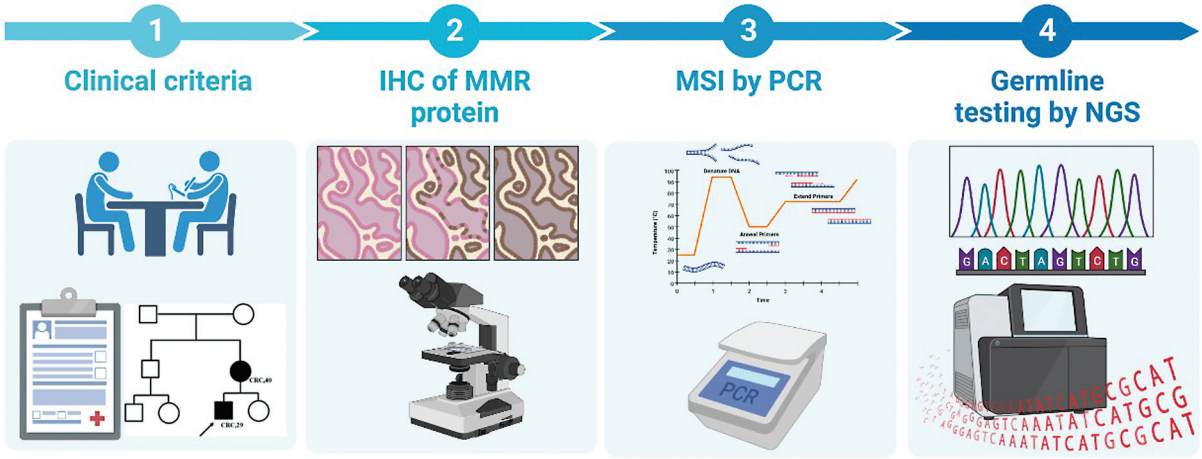

Fig. 1 Evolution of Lynch syndrome (LS) screening in China (created with BioRender.com). IHC, immunohistochemistry; MMR, mismatch pairing; MSI, microsatellite instability; NGS, next-generation sequencing; PCR, polymerase chain reaction.

Table 2 Different clinical diagnostic criterion of Lynch syndrome

Criteria	Pedigree information	Additional criteria
Amsterdam criteria I	At least 3 relatives with CRC	All the following criteria should be present: (1) one should be a first-degree relative of the other two; (2) at least two successive generations should be affected; (3) at least one CRC should be diagnosed before age 50; (4) FAP should be excluded
Amsterdam criteria II	At least 3 relatives with LS-associated cancer	All the following criteria should be present: (1) one should be a first-degree relative of the other two; (2) at least two successive generations should be affected; (3) at least one CRC should be diagnosed before age 50; (4) FAP should be excluded
Chinese criteria for LS	At least 2 siblings affected by CRC in a family	Any of the following criteria is present: (1) development of multiple CRC; (2) at least one CRC diagnosed before the age of 50 years; (3) development of LS-associated extracolonic cancer in family

Abbreviations: CRC, colorectal cancer; FAP, familial adenomatous polyposis; LS, Lynch syndrome.

combines with the microsatellite status. Though these guidelines bring the sensitivity to a higher level, they are complicated and multiple factors are involved, resulting in a low specificity, which makes it less clinically practical. Efforts have been made to improve the clinical application of those guidelines utilizing computer methods,[35–37] but they seemed to be unavailing since those models were more likely to identified highest-risk individuals rather than the whole group of LS patients.[38] Therefore, in Sun Yat-sen University Cancer Center (SYSUCC), China, only patients fulfilling the Bethesda guidelines but with MMR proficiency (pMMR) CRC are recommended to perform microsatellite instability (MSI) assay for verification, aiming to identify real susceptive individuals who need further genetic examination.

Molecular Testing

With the development of molecular methods, the strategy called universal screening provides a powerful tool for LS screening, based on MMR protein immunohistochemistry (IHC) and deoxyribonucleic acid (DNA) MSI testing, and its feasibility and effectiveness have been verified by several studies.[19,39,40] In this strategy, MMR protein IHC or MSI test will be performed in tumor of CRC patient and if either test is positive, further MMR genes germline test will be performed to confirm the diagnosis of LS after excluding the hypermethylation of MLH1 promoter region.

The two assays mentioned above can be used independently or together though their combination was able to maximize the specificity and sensitivity of identifying LS individual.[41–43] However, the prevalence of LS varies in different countries[4,18,20,44] and ethnic heterogeneity may hamper the universal screening to be really "universal" worldwide. A thorough understanding of the prevalence and genetic mutation characteristics of LS in a certain region helps to identify the candidates for further germline sequencing and optimize the screening strategy. A single-institute study on a large cohort from SYSUCC showed a

Table 3 Studies on the prevalence of dMMR

Reference/Nationality	No. of cases	Incidence of dMMR
Hampel et al[19] (USA)	1,566	14.7%
Heald et al[40] (USA)	1,108	16.0%
Jiang et al[21] (China)	3,250	10.2%
Yang et al[22] (China)	4,966	6.0%

Abbreviation: dMMR, mismatch repair deficiency.

Table 4 Studies on the prevalence of BRAFV600E mutation in dMLH1

Reference/Nationality	No. of cases	Incidence of BRAF mutation in dMLH1
Ward et al[6] (Australia)	205	75.0%
Chika et al[44] (Japan)	54	51.9%
Adar et al[39] (USA)	126	69.0%
Jiang et al[21] (China)	154	9.7%

Abbreviation: dMLH1, MLH1 deficiency.

different picture of the application of universal screening in Chinese population.[21] In this study, universal screening was conducted in a consecutive cohort with newly diagnosed CRC, using IHC for MMR proteins, followed by BRAFV600E testing in cases with absence of MLH1, and then multigene panel testing on germline DNA in all cases with MMR deficiency (dMMR) and no BRAFV600E mutation. The incidence of dMMR was 10.2% in these 3,250 patients, which was modestly lower than that of the American population[19,40] (►**Table 3**). Moreover, the prevalence of LS was 2.9% in this cohort, which was similar to the population studies of Salovaara et al[18] and Hampel et al[19] but much higher than that in Spain and Japan.[20,44] Notably, in this study, only 9.7% of dMLH1 patients carried BRAFV600E mutation, which was remarkably less than that in previous studies[6,39,44] (►**Table 4**). And one-third of the pathogenic or likely pathogenic variants (PV/LPV) have not been reported previously, suggesting there was a different mutation spectrum in this population and the universal screening strategy may not fit perfectly, which was further verified by the relatively low positive predictive value (36.3%) in this cohort. Due to the low risk (5.7%) of LS in patients older than 65 years with dMMR CRC in this study, a selective strategy was proposed: only patients with dMMR tumors and an onset age of 65 years or below, and older patients fulfilling at least one criterion of the revised Bethesda guidelines were needed to perform germline sequencing. In this case, 8.2% fewer cases would be candidates for germline sequencing while none of the LS individuals being omitted and positive predictive value of the strategy was slightly higher than that of universal screening (►**Table 5**). Considering the low incidence of BRAFV600E mutation and the low prevalence of LS in older patients in Chinese population, this selective strategy was optimal and could be an alternative approach to universal screening for LS, especially in Chinese population. On the basis of this comprehensive single-institute study on Chinese population, a multicenter study was conducted by Yang et al,[22] aiming to further reveal the genetic spectrum of dMMR CRC and to develop a nomogram for screening LS in China. The IHC results of the four MMR genes in 4,966 postoperative patients from 15 hospitals across China were examined and the prevalence of dMMR was 6.2%, which provided more solid evidence for the lower incidence of dMMR in Chinese population. Among 311 enrolled dMMR patients, 95 (30.5%) of them harbored germline PV/LPV in MMR genes and were diagnosed with LS consequently. Then, clinical manifestations were compared between the LS group and sporadic group and a nomogram was developed based on these clinical characteristics differences including age of onset, sex, personal history, family history of first- and second-degree relatives, and the patents of dMMR. This nomogram was efficient in classifying LS and non-LS associated dMMR, with an area under curve (AUC) of 0.87, higher than the AUC of other screening strategies like the Amsterdam criteria II, Bethesda criteria, Chinese criteria for LS, as well as the selective strategy, and was validated in another external cohort of Chinese population.[21] Furthermore, according to the nomogram, patients with a LS probability > 0.435 were recommended to take germline assays and genetic counseling, as this cutoff value achieves a specificity of 0.889 and a sensitivity of 0.716. In China, the screening of LS is still under development, thus this Chinese

Table 5 Performance of different strategies for the identification of patients with Lynch syndrome[21]

Screening strategy	Case (%)		Diagnostic yield (%)	Sensitivity (%)	PPV (%)
	Patients requiring MMR testing	Patients requiring germline sequencing			
Amsterdam criteria II	0 (0)	35 (1.1)	0.3	9.7	25.7
Revised Bethesda guidelines	1,046 (32.7)	164 (5.1)	2.4	81.7	7.3
Universal screening	3,191 (100)	256 (8.0)	2.9	100	36.3
Selective strategy	3,191 (100)	235 (7.4)	2.9	100	39.6

Abbreviations: MMR, mismatch repairing; PPV, positive predictive value.

Table 6 Studies on the patents of dMMR

Reference/Nationality	No. of cases	Incidence and patents of dMMR				
		dMLH1 alone or with partner	dMSH2 alone or with partner	Isolated dMSH6	Isolated dPMS2	Other
Ward et al[6] (Australia)	245	83.7%	7.8%	4.9%	2.0%	0.8%
Pérez-Carbonell et al[20] (Spain)	155	74.2%	18.7%	5.8%	1.3%	/
Jiang et al[21] (China)	330	47.9%	30.0%	7.9%	7.9%	6.7%
Yang et al[22] (China)	311	59.5%	22.5%	6.4%	11.6%	/

Abbreviations: dMLH1, MLH1 deficiency; dMMR, mismatch repair deficiency.

data-based accessible predictive model has great clinical promotion value considering the declining family size and distinct genetic landscape in China. According to this result, a prospective real-world study aiming to further validate and optimize the effectiveness of this nomogram is ongoing in several medical centers in China.

Besides the occurrence of dMMR, studies by Jiang et al[21] and Yang et al[22] also revealed the diversity of the patents of dMMR. The incidence of MLH1 deficiency (dMLH1) alone or with its partner PMS2 in these two studies was 47.9 and 59.5%, respectively, remarkably lower than that in Australian (83.7%)[6] and Spanish (74.5%)[20] population (►**Table 6**). This mutation diversity prompts the screening strategy to be modified. Given that the incidence of dMLH1 and BRAFV600E mutation is lower in Chinese population, the efficiency of different screening strategies for LS, including BRAF testing and MLH1 methylation testing alone or their combination, and the combination of revised Bethesda criteria and MLH1 methylation testing in Chinese patients with dMLH1 CRC was compared by Xiao et al.[45] Among the 109 eligible cases, even the combination of BRAF testing and MLH1 methylation testing showed poor performance with a specificity of 47.7% and this indicated that BRAF testing and MLH1 promoter methylation testing as prescreens to exclude LS were less effective in Chinese CRC patients than in western CRC patients.[46,47] Consequently, in view of the low frequency of dMLH1 and BRAFV600E variants in China, for Chinese population with dMMR CRC, direct germline test rather than BRAF examination is preferred, due to its higher sensitivity, specificity, and more importantly, higher cost performance.[45]

Since NGS germline examination with multigene panels has become a crucial method for detecting individuals at risk of hereditary disease (►**Table 7**), Jiang et al[2] further explored the clinical application of this method by conducting a prospective study among patients with CRC. The incidence of germline PV was 7.8% in all 486 eligible patients where LS was identified in 20 patients (4.1%), using a comprehensive commercial panel which comprised 81 genes. All the clinically relevant PVs were found in patients diagnosed under age 70 years while patients carrying PVs in genes from the additional testing set were older than 40 years, indicating universal germline testing for cancer susceptibility genes should be recommended among all patients with CRC diagnosed under age 70 years and a broad panel including genes from the additional testing set might be considered for patients with CRC older than 40 years to clarify inheritance risks. Although universal germline testing is able to identify LS individuals presenting with pMMR tumors, it is worth noting that the wide application of germline testing as well as the broaden panel may increase the complexities of genetic risk assessment and the cost of clinical care in real-world setting.

Technique Improvement

The detection of the loss expression of MMR protein in CRC tumor through IHC is nowadays a common approach for LS screening. However, IHC assay is relatively costly and the accurate evaluation is highly dependent on experienced pathologists, thus making it less accessible in underdeveloped countries.[13] The result of a retrospective study on T4bm0 CRC from SYSUCC indicated that the incidence of dMMR in T4bM0 CRC (25.0%) was significantly higher than that in unselected CRC population. Additionally, patients in this group of patients showed distinct clinical manifestations such as tumor location, tumor size, family history, and pathological type, which can help to better identify patients with dMMR CRC in clinical care. In addition to clinical phenotypes, the rapid development of artificial intelligent and deep learning image analysis show promising application prospect in histopathological evaluation[48,49] in the past decade and make it possible to predict CRC MMR status from easily accessible hematoxylin and eosin-stained whole-slide images (WSIs) through a deep learning-based method. Jiang et al developed a multiple instance learning model to predict MMR status in CRC specimen. Their model achieved an AUC of 0.8888 in 441 WSIs from the TCGA-validation cohort and the efficiency was further validated in external cohorts including Pathology AI Platform (AUC = 0.8806), SYSUCC-surgical cohort (AUC = 0.8457), and even a biopsy specimen cohort (AUC = 0.7679). Furthermore, a dual-threshold triage strategy was then built with a sensitivity higher than 90% and a specificity higher than 95% to minimize more than 50% of patients avoiding IHC-based MMR testing, indicating that it had excellent performance and would be a great supplement to the current screening startegy.[50]

Microsatellite sequences are short-repeated DNA sequences whose lengths are 1 to 6 base pair(s) in the genome. Polymerase chain reaction (PCR) assay was widely used to

Table 7 Studies on multigene panel testing in Lynch syndrome/hereditary CRC

Reference	Panel type	Study population	Key germline findings
High-risk patient cohort			
Yurgelun et al 2015[64]	Commercial 25-gene panel	Laboratory-based cohort of 1,260 patients referred for LS germline testing	14.4% prevalence of any mutation; 9.0% LS prevalence
Espenschied et al[65]	Various commercial panels (9–49 genes)	Laboratory-based cohort of 34,981 patients referred for multigene panel germline testing for a variety of clinical indications	Overall 1.5% LS prevalence; MSH6 mutations most common among carriers with ovarian or endometrial cancer; PMS2 mutations most common among carriers with breast cancer only
Cohort of selected patients with CRC			
Jiang et al[2]	Commercial 81-gene panel (minimal set, additional set and others)	Multisite, population-based cohort of 486 patients with CRC aged $\leq$ 70 years or aged $>$ 70 years but fulfilled the revised Bethesda guidelines or polyposis syndromes diagnostic criteria	7.8% prevalence of any mutation; 4.1% LS prevalence; All mutations occurred in patients aged under 70 years while patients with mutation in the additional set were older than 40 years
Cohorts of patients with CRC not preselected by personal or family history of cancer			
Yurgelun et al 2017[1]	Commercial 25-gene panel	Single-site, clinic-based cohort of 1,058 patients with CRC	9.9% prevalence of any mutation; 3.1% LS prevalence
Pearlman et al[66]	Commercial 25-gene panel	Multisite, population-based cohort of 450 patients with CRC aged $<$ 50 years	16.0% prevalence of any mutation; 8.4% LS prevalence

Abbreviations: CRC, colorectal cancer; LS, Lynch syndrome.

evaluate the MSI status. The sensitivity of MSI assay detection for LS carriers ranges from 55 to 91%.[51] Nowadays, NGS has gradually become a standard method to evaluate MSI with a high validity. MSI test performed by NGS greatly enhances LS detection beyond conventional MSI/IHC.[52,53] Besides, NGS provides a more comprehensive information of genes variants, far more than the four MMR genes. MSIsensor[52] and mSINGS,[54] for example, are two major approaches to examine the MSI status by measuring the read count distribution directly, and both of them have achieved favorable sensitivity and specificity for LS carrier screening. MSI-ColonCore established by Zhu et al was also a read-count–distribution-based method using the ColonCore panel, an NGS panel designed to detect MSI status and variants in various CRC-related genes.[55] Comparing to the gold standard MSI-PCR test, MSI-ColonCore achieved a high sensitivity of 97.9% and a specificity of 100% for the detection of MSI status. Additionally, MSI-ColonCore also showed more efficient and robust performance compared with MSIsensor and mSINGS, indicating its reliable clinical applicability.

Although the combination of IHC and MSI assay can greatly improve the performance in screening candidates for germline testing, the discordance of these two results can be confusing and misleading. Given that the evaluation of the MMR protein expression by IHC is highly dependent on the experience of pathologists, it is reported that the IHC MMR expression results may vary from different institutes.[56] The deep-learning-based evaluation method developed by Jiang et al can overcome this shortage to a great extent. In this scenario, the combination of new technique-based IHC and MSI assay will be more precise in LS screening and diagnosis.

Management of LS Individuals

As the screening strategies for LS have been continuously optimized, clinical geneticists shed more light on the management of LS individuals and their families. Of concern, although more attention to hereditary diseases including LS has been paid and the development of biological molecular technique makes the genetic testing more accessible, non-indicated testing should be avoided to reduce the burden of health care. In view of this, rapid and cost-effective identification of individuals at risks of hereditary diseases can tremendously help to manage the referral to genetic assay, which is of crucial significance. Several occidental studies have focused on utilizing online questionnaires to collect personal and family medical history so as to better identify patients at risks,[57,58] but their results were not so encouraging, which may result from the relatively well-developed genetic counseling system in western countries. But in China, this system is immature since qualified genetics professionals are rare and concentrated in a few tertiary referral hospitals, making it less accessible for risk evaluation nationwide. Therefore, convenient online risk assessment tools

are in great demand for Chinese population. Several electronic tools have been established via mobile software. After submitting personal and family medical history, estimated risk of LS will present with the corresponding medical suggestions including referral to genetic counseling and more frequent surveillance for LS-associated cancers. And effectiveness of these tools is under investigation. Besides risk assessment, efforts also have been made in China to explore other potential genome variants that may lead to LS, except for the four canonical MMR genes, and variants of unknown significance (VUS), which can provide more comprehensive evidence of this hereditary syndrome and help improve the screening strategy, especially when the mutation spectrum of LS is distinct in Chinese population, as mentioned above.[21] MLH3 mutations seems to be low-risk factor for LS since mutations of this gene alone may not impair the MMR function[59] and its deficiency can play a functional role in tumorigenicity via its interaction with other MMR genes like MLH1 and PMS2.[60,61] Sui et al identified a germline MLH1 VUS and a novel germline MLH3 mutation in a Chinese LS patient.[62] Although the MLH3 variant was not detected among other patients of the maternal pedigree, it may have enhanced tumorigenicity in this case, owing to the younger onset age of the proband than the other patients only carrying MLH1 variant in his family. The significance of MLH3 in LS was further investigated by Dr. Yang. They reported a family suspected with LS where the proband was a homozygous carrier of the MLH3 truncating variant but they supposed that the biallelic germline frameshift variant they found was not the pathogenic defect after analyzing the clinical features and inheritance pattern of this pedigree. The impact of MLH3 mutations in LS requires deeper exploration. In addition, a national database (http://cfcsg-database.org.cn/) has been built up to record germline mutations of hereditary cancers in China, aiming to accumulate more information and provide solid evidence for the improvement of screening strategies and treatment.

Outlook and Summary

With the declining cost and consequent wide application of gene mutation examination, the screening and diagnostics of LS and other hereditary CRC have achieved remarkable progress, implying a favorable development of precision medicine. However, there are several issues demanding exploration, such as Lynch-like syndrome (LLS). Patients with dMMR/MSI-H CRC without germline pathogenic or suspected pathogenic variants of the MMR gene after exclusion of hypermethylation in the MLH1 promoter or BRAF mutations are generally considered to be LLS, which was reported to account for up to 30% of the dMMR/MSI-H CRC.[20] And the application of advanced technique like long-read sequencing may contribute to better separation from LS and LLS.[63] What's more, the screening strategy for LS has been validated in CRC and efforts should be made to take this tactic to other LS-associated cancers.

The variation information provided by bioinformatic analysis can help better identify individuals with genetic risks, and all the mutation information as well as their corresponding unique molecular phenotypes will provide more comprehensive understanding of the ethnic heterogeneity. In China, screening strategies for LS has been continuously modified relying on the local evidence. There are several distinctions in Chinese LS individuals and families: the family size is minor; the incidence of dMMR and dMLH1 is lower than that in other countries; and the prevalence of BRAFV600E mutation in dMMR is also lower. All these features prompt the clinical geneticists to optimize the screening strategy and now they are making good progress. Moreover, Chinese experience not only greatly contributes to a comprehensive understanding of LS, but also brings an applicable nomogram and deep learning-based MMR status-predicting tool into practice. As molecular technique continues to develop and the accumulation of global experience grows, it is crucial to further understand the ethnic heterogeneity and optimize the screening strategy.

Funding
This study was funded by the National Natural Science Foundation of China (grant numbers 82073159 and 81871971) and Guangdong Basic and Applied Basic Research Foundation (2022A1515012403).

Conflict of Interest
None declared.

References

1 Yurgelun MB, Kulke MH, Fuchs CS, et al. Cancer susceptibility gene mutations in individuals with colorectal cancer. J Clin Oncol 2017; 35(10):1086–1095
2 Jiang W, Li L, Ke CF, et al. Universal germline testing among patients with colorectal cancer: clinical actionability and optimised panel. J Med Genet 2022;59(04):370–376
3 Aaltonen LA, Salovaara R, Kristo P, et al. Incidence of hereditary nonpolyposis colorectal cancer and the feasibility of molecular screening for the disease. N Engl J Med 1998;338(21):1481–1487
4 Hampel H, Frankel WL, Martin E, et al. Screening for the Lynch syndrome (hereditary nonpolyposis colorectal cancer). N Engl J Med 2005;352(18):1851–1860
5 Moreira L, Balaguer F, Lindor N, et al; EPICOLON Consortium. Identification of Lynch syndrome among patients with colorectal cancer. JAMA 2012;308(15):1555–1565
6 Ward RL, Hicks S, Hawkins NJ. Population-based molecular screening for Lynch syndrome: implications for personalized medicine. J Clin Oncol 2013;31(20):2554–2562
7 Win AK, Young JP, Lindor NM, et al. Colorectal and other cancer risks for carriers and noncarriers from families with a DNA mismatch repair gene mutation: a prospective cohort study. J Clin Oncol 2012;30(09):958–964
8 Bonadona V, Bonaïti B, Olschwang S, et al; French Cancer Genetics Network. Cancer risks associated with germline mutations in MLH1, MSH2, and MSH6 genes in Lynch syndrome. JAMA 2011; 305(22):2304–2310
9 Watson P, Vasen HFA, Mecklin JP, et al. The risk of extra-colonic, extra-endometrial cancer in the Lynch syndrome. Int J Cancer 2008;123(02):444–449
10 Vasen HF, Watson P, Mecklin JP, Lynch HT. New clinical criteria for hereditary nonpolyposis colorectal cancer (HNPCC, Lynch syndrome) proposed by the International Collaborative group on HNPCC. Gastroenterology 1999;116(06):1453–1456

11 Rodriguez-Bigas MA, Boland CR, Hamilton SR, et al. A National Cancer Institute Workshop on Hereditary Nonpolyposis Colorectal Cancer Syndrome: meeting highlights and Bethesda guidelines. J Natl Cancer Inst 1997;89(23):1758–1762
12 Umar A, Boland CR, Terdiman JP, et al. Revised Bethesda Guidelines for hereditary nonpolyposis colorectal cancer (Lynch syndrome) and microsatellite instability. J Natl Cancer Inst 2004;96 (04):261–268
13 Shia J. The diversity of tumours with microsatellite instability: molecular mechanisms and impact upon microsatellite instability testing and mismatch repair protein immunohistochemistry. Histopathology 2021;78(04):485–497
14 Win AK, Jenkins MA, Dowty JG, et al. Prevalence and penetrance of major genes and polygenes for colorectal cancer. Cancer Epidemiol Biomarkers Prev 2017;26(03):404–412
15 Goodenberger ML, Thomas BC, Riegert-Johnson D, et al. PMS2 monoallelic mutation carriers: the known unknown. Genet Med 2016;18(01):13–19
16 ten Broeke SW, Brohet RM, Tops CM, et al. Lynch syndrome caused by germline PMS2 mutations: delineating the cancer risk. J Clin Oncol 2015;33(04):319–325
17 Ryan NAJ, Morris J, Green K, et al. Association of mismatch repair mutation with age at cancer onset in Lynch syndrome: implications for stratified surveillance strategies. JAMA Oncol 2017;3 (12):1702–1706
18 Salovaara R, Loukola A, Kristo P, et al. Population-based molecular detection of hereditary nonpolyposis colorectal cancer. J Clin Oncol 2000;18(11):2193–2200
19 Hampel H, Frankel WL, Martin E, et al. Feasibility of screening for Lynch syndrome among patients with colorectal cancer. J Clin Oncol 2008;26(35):5783–5788
20 Pérez-Carbonell L, Ruiz-Ponte C, Guarinos C, et al. Comparison between universal molecular screening for Lynch syndrome and revised Bethesda guidelines in a large population-based cohort of patients with colorectal cancer. Gut 2012;61(06):865–872
21 Jiang W, Cai MY, Li SY, et al; Written on behalf of AME Colorectal Cancer Cooperative Group. Universal screening for Lynch syndrome in a large consecutive cohort of Chinese colorectal cancer patients: High prevalence and unique molecular features. Int J Cancer 2019;144(09):2161–2168
22 Yang M, Li D, Jiang W, et al. Development and external validation of a novel nomogram for screening Chinese Lynch syndrome: based on a multicenter, population study. Ther Adv Med Oncol 2021;13(Jun):17588359211023290
23 Vasen HF. Clinical description of the Lynch syndrome [hereditary nonpolyposis colorectal cancer (HNPCC)]. [hereditary nonpolyposis colorectal cancer (HNPCC)]Fam Cancer 2005;4(03): 219–225
24 Perea J, Rodríguez Y, Rueda D, et al. Early-onset colorectal cancer is an easy and effective tool to identify retrospectively Lynch syndrome. Ann Surg Oncol 2011;18(12):3285–3291
25 Syngal S, Brand RE, Church JM, Giardiello FM, Hampel HL, Burt RWAmerican College of Gastroenterology. ACG clinical guideline: genetic testing and management of hereditary gastrointestinal cancer syndromes. Am J Gastroenterol 2015;110(02):223–262, quiz 263
26 Giardiello FM, Allen JI, Axilbund JE, et al; US Multi-Society Task Force on Colorectal Cancer. Guidelines on genetic evaluation and management of Lynch syndrome: a consensus statement by the US Multi-Society Task Force on colorectal cancer. Gastroenterology 2014;147(02):502–526
27 de Vos tot Nederveen Cappel WH, Buskens E, van Duijvendijk P, et al. Decision analysis in the surgical treatment of colorectal cancer due to a mismatch repair gene defect. Gut 2003;52(12): 1752–1755
28 Vasen HF, Nagengast FM, Khan PM. Interval cancers in hereditary non-polyposis colorectal cancer (Lynch syndrome). Lancet 1995; 345(8958):1183–1184
29 Jenkins MA, Hayashi S, O'Shea AM, et al; Colon Cancer Family Registry. Pathology features in Bethesda guidelines predict colorectal cancer microsatellite instability: a population-based study. Gastroenterology 2007;133(01):48–56
30 Bessa X, Alenda C, Paya A, et al. Validation microsatellite path score in a population-based cohort of patients with colorectal cancer. J Clin Oncol 2011;29(25):3374–3380
31 Vasen HF, Mecklin JP, Khan PM, Lynch HT. The International Collaborative Group on Hereditary Non-Polyposis Colorectal Cancer (ICG-HNPCC). Dis Colon Rectum 1991;34(05):424–425
32 Barnetson RA, Tenesa A, Farrington SM, et al. Identification and survival of carriers of mutations in DNA mismatch-repair genes in colon cancer. N Engl J Med 2006;354(26):2751–2763
33 Yuan Y, Han HJ, Zheng S, Park JG. Germline mutations of hMLH1 and hMSH2 genes in patients with suspected hereditary nonpolyposis colorectal cancer and sporadic early-onset colorectal cancer. Dis Colon Rectum 1998;41(04):434–440
34 Yuan Y, Ye J, Zheng SInternational Collaborative Group. Clinical and genetic features of International Collaborative Group-hereditary nonpolyposis colorectal cancer families and suspected hereditary nonpolyposis colorectal cancer families. Chin Med J (Engl) 2004;117(05):748–752
35 Chen S, Wang W, Lee S, et al; Colon Cancer Family Registry. Prediction of germline mutations and cancer risk in the Lynch syndrome. JAMA 2006;296(12):1479–1487
36 Kastrinos F, Steyerberg EW, Balmaña J, et al; Colon Cancer Family Registry. Comparison of the clinical prediction model PREMM (1,2,6) and molecular testing for the systematic identification of Lynch syndrome in colorectal cancer. Gut 2013;62(02):272–279
37 Green RC, Parfrey PS, Woods MO, Younghusband HB. Prediction of Lynch syndrome in consecutive patients with colorectal cancer. J Natl Cancer Inst 2009;101(05):331–340
38 Kastrinos F, Ojha RP, Leenen C, et al; Lynch Syndrome prediction model validation study group. Comparison of prediction models for Lynch syndrome among individuals with colorectal cancer. J Natl Cancer Inst 2015;108(02):djv308
39 Adar T, Rodgers LH, Shannon KM, et al. Universal screening of both endometrial and colon cancers increases the detection of Lynch syndrome. Cancer 2018;124(15):3145–3153
40 Heald B, Plesec T, Liu X, et al. Implementation of universal microsatellite instability and immunohistochemistry screening for diagnosing lynch syndrome in a large academic medical center. J Clin Oncol 2013;31(10):1336–1340
41 Shia J. Immunohistochemistry versus microsatellite instability testing for screening colorectal cancer patients at risk for hereditary nonpolyposis colorectal cancer syndrome. Part I. The utility of immunohistochemistry. J Mol Diagn 2008;10(04):293–300
42 Zhang L. Immunohistochemistry versus microsatellite instability testing for screening colorectal cancer patients at risk for hereditary nonpolyposis colorectal cancer syndrome. Part II. The utility of microsatellite instability testing. J Mol Diagn 2008;10(04): 301–307
43 Cohen SA, Pritchard CC, Jarvik GP. Lynch syndrome: from screening to diagnosis to treatment in the era of modern molecular oncology. Annu Rev Genomics Hum Genet 2019;20:293–307
44 Chika N, Eguchi H, Kumamoto K, et al. Prevalence of Lynch syndrome and Lynch-like syndrome among patients with colorectal cancer in a Japanese hospital-based population. Jpn J Clin Oncol 2017;47(02):108–117
45 Xiao B, Luo J, Xie E, et al. Comparisons of screening strategies for identifying Lynch syndrome among patients with MLH1-deficient colorectal cancer. Eur J Hum Genet 2020;28(11):1555–1562
46 Adar T, Rodgers LH, Shannon KM, et al. A tailored approach to BRAF and MLH1 methylation testing in a universal screening program for Lynch syndrome. Mod Pathol 2017;30(03):440–447
47 Moreira L, Muñoz J, Cuatrecasas M, et al; Gastrointestinal Oncology Group of the Spanish Gastroenterological Association. Prevalence of somatic mutl homolog 1 promoter hypermethylation in

Lynch syndrome colorectal cancer. Cancer 2015;121(09): 1395–1404
48 Jiang Y, Yang M, Wang S, Li X, Sun Y. Emerging role of deep learning-based artificial intelligence in tumor pathology. Cancer Commun (Lond) 2020;40(04):154–166
49 Mori Y, Bretthauer M, Kalager M. Hopes and hypes for artificial intelligence in colorectal cancer screening. Gastroenterology 2021;161(03):774–777
50 Jiang W, Mei WJ, Xu SY, et al. Clinical actionability of triaging DNA mismatch repair deficient colorectal cancer from biopsy samples using deep learning. EBioMedicine 2022;81(Jul):104120
51 Evaluation of Genomic Applications in Practice and Prevention (EGAPP) Working Group. Recommendations from the EGAPP Working Group: genetic testing strategies in newly diagnosed individuals with colorectal cancer aimed at reducing morbidity and mortality from Lynch syndrome in relatives. Genet Med 2009; 11(01):35–41
52 Benson AB, Venook AP, Al-Hawary MM, et al. NCCN Guidelines Insights: Colon Cancer, Version 2.2018. J Natl Compr Canc Netw 2018;16(04):359–369
53 Niu B, Ye K, Zhang Q, et al. MSIsensor: microsatellite instability detection using paired tumor-normal sequence data. Bioinformatics 2014;30(07):1015–1016
54 Salipante SJ, Scroggins SM, Hampel HL, Turner EH, Pritchard CC. Microsatellite instability detection by next generation sequencing. Clin Chem 2014;60(09):1192–1199
55 Zhu L, Huang Y, Fang X, et al. A novel and reliable method to detect microsatellite instability in colorectal cancer by next-generation sequencing. J Mol Diagn 2018;20(02):225–231
56 Jiang W, Sui QQ, Li WL, et al. Low prevalence of mismatch repair deficiency in Chinese colorectal cancers: a multicenter study. Gastroenterol Rep (Oxf) 2020;8(05):399–403
57 Kallenberg FG, IJspeert JE, Bossuyt PM, Aalfs CM, Dekker E. Validation of an online questionnaire for identifying people at risk of familial and hereditary colorectal cancer. Fam Cancer 2015; 14(03):401–410
58 Kallenberg FGJ, Aalfs CM, The FO, et al. Evaluation of an online family history tool for identifying hereditary and familial colorectal cancer. Fam Cancer 2018;17(03):371–380
59 Korhonen MK, Vuorenmaa E, Nyström M. The first functional study of MLH3 mutations found in cancer patients. Genes Chromosomes Cancer 2008;47(09):803–809
60 Chen PC, Dudley S, Hagen W, et al. Contributions by MutL homologues Mlh3 and Pms2 to DNA mismatch repair and tumor suppression in the mouse. Cancer Res 2005;65(19):8662–8670
61 Chen PC, Kuraguchi M, Velasquez J, et al. Novel roles for MLH3 deficiency and TLE6-like amplification in DNA mismatch repair-deficient gastrointestinal tumorigenesis and progression. PLoS Genet 2008;4(06):e1000092
62 Sui QQ, Jiang W, Wu XD, Ling YH, Pan ZZ, Ding PR. A frameshift mutation in exon 19 of MLH1 in a Chinese Lynch syndrome family: a pedigree study. J Zhejiang Univ Sci B 2019;20(01):105–108
63 Te Paske IBAW, Mensenkamp AR, Neveling K, Hoogerbrugge N, Ligtenberg MJL, De Voer RMERN-GENTURIS Lynch-Like Working Group. Noncoding aberrations in mismatch repair genes underlie a substantial part of the missing heritability in Lynch syndrome. Gastroenterology 2022;163(06):1691–1694.e7
64 Yurgelun MB, Allen B, Kaldate RR, et al. Identification of a variety of mutations in cancer predisposition genes in patients with suspected Lynch syndrome. Gastroenterology 2015;149(03): 604–13.e20
65 Espenschied CR, LaDuca H, Li S, McFarland R, Gau CL, Hampel H. Multigene panel testing provides a new perspective on Lynch syndrome. J Clin Oncol 2017;35(22):2568–2575
66 Pearlman R, Frankel WL, Swanson B, et al; Ohio Colorectal Cancer Prevention Initiative Study Group. Prevalence and spectrum of germline cancer susceptibility gene mutations among patients with early-onset colorectal cancer. JAMA Oncol 2017;3(04):464–471