Comprehensive review of mouse models for studying cancer-related fatigue: Methods, findings and future directions (Review)

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Abstract. Cancer-related fatigue (CRF) is one of the most prevalent and debilitating symptoms experienced by patients with cancer, significantly impacting their quality of life. Currently, the precise mechanisms underlying the onset of this fatigue remain poorly understood, and there is a notable lack of effective pharmacological treatments to alleviate it. Therefore, establishing a reliable and stable mouse model of CRF is essential for advancing research in this important area. The present study systematically reviewed various methods for inducing CRF in mice, including tumor-bearing models, chemotherapy and localized irradiation, either used alone or in combination. The advantages and disadvantages of each of these models were analyzed, providing researchers with valuable insights and references for selecting the most appropriate mouse model of CRF for their specific studies.

Contents

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1. Introduction

Cancer-related fatigue (CRF) is a form of physical, emotional and/or cognitive fatigue associated with cancer itself or its treatment. It is a remarkably distressing subjective sensation that persists for an extended duration, disproportionate to recent activity, and significantly interferes with normal functioning, thereby impacting patients' quality of life (1,2). CRF is prevalent among patients with cancer, with nearly all patients reporting symptoms of fatigue (3,4). Currently, the treatment of CRF primarily involves non-specific medications (such as antidepressants, analgesics and stimulants), psychological interventions (such as cognitive behavioral therapy) and physical activity interventions. However, due to the unclear pathophysiology of CRF, these symptomatic treatments are of limited effectiveness or are accompanied by side effects.

Mice are commonly used in experimental studies of CRF, and the establishment of mouse models that accurately simulate the fatigue symptoms experienced by patients with cancer is crucial for investigating the mechanisms of CRF. Currently, several researchers have developed various mouse models of CRF using different approaches. However, there is inconsistency across these models in terms of strain selection, modeling methods and fatigue assessment criteria, and standardized protocols have yet to be established. Furthermore, although technologies such as metabolomics and proteomics have made significant progress in exploring the mechanisms of CRF, specific therapeutic targets for CRF remain unclear. A substantial gap still exists between clinical research and experimental studies. The present review summarizes and discusses the modeling methods for CRF in mice and the underlying mechanisms of fatigue, with the aim of providing insights for further research on CRF.

2. Developing a CRF model using tumor-bearing mice

Colon cancer-induced CRF mouse models. The Colon Tumor 26 (CT26) cell line is a commonly used mouse colon cancer cell line that is characterized by rapid proliferation and the ability to form both solid and metastatic tumors. Pin *et al* (5) subcutaneously implanted $1x10^6$ CT26 cells into the dorsal area of male CD2F1 mice, while Peters *et al* (6) performed

subcutaneous implantation of the same number of CT26 cells in the inguinal region. Shen *et al* (7) conducted a similar experiment but selected BALB/c mice instead. These researchers successfully established cancer-induced fatigue models, with experimental results showing that CT26 tumor-bearing mice exhibited a decrease in both body weight and tumor-free body weight, reduced skeletal muscle mass and strength, and significant fatigue symptoms. Other researchers used higher doses of CT26 cells for subcutaneous implantation, with several researchers subcutaneously implanting $5x10^6$ CT26 cells into different mouse strains to establish tumor-bearing mouse fatigue models (8-13). For example, Roberts *et al* (12) and Murphy *et al* (10) used male CD2F1 mice, while Lee *et al* (9) and Song *et al* (13) used male BALB/c mice, and Norden *et al* (11) and Clark *et al* (8) conducted experiments with female CD2F1 mice. Norden *et al* (11,14) suggested that female mice bearing tumors could better maintain food intake, exhibit less weight loss and reduce the likelihood of local inflammation caused by mice gnawing at the tumor site, making them more suitable for studying cancer-induced fatigue.

In addition to subcutaneous implantation, intravenous injection of CT26 cells can also be used to establish a tumor-bearing mouse model. Ferdaos *et al* (15) injected $5x10^6$ CT26 cells intravenously into one group of BALB/c mice, while another group received $2x10^6$ CT26 cells subcutaneously in the back. Both groups exhibited fatigue symptoms and lung metastases, with a comparable tumor count. Compared with mice subjected to subcutaneous or intravenous injection of tumor cells, colon orthotopic tumor-bearing mice may simulate more closely the real conditions of patients with colon cancer. Zhang *et al* (16) reported a method for colon cancer orthotopic tumor modeling: Tumors formed by subcutaneously implanted CT26 cells were cut into 2x2x1 mm pieces and surgically implanted in the cecum of mice, with a success rate of 62.5%. Orthotopic tumors may better mimic the clinical scenario of colon cancer, but the procedure is more complex and has a lower success rate. Additionally, ApcMin/+(MIN) mice carrying a mutant Apc gene can also be used for colon cancer research. VanderVeen *et al* (17) found that male C57BL/6J MIN mice exhibited a 6% weight loss at 18 weeks of age, with an 8% reduction in forelimb grip strength and tibialis anterior muscle mass, and fatigue symptoms appeared before significant weight loss. Hetzler *et al* (18) observed that these mice showed sex differences, with female MIN mice experiencing less weight loss and lower blood IL-6 levels. This mouse model does not require tumor cell implantation, thus offering improved experimental stability; however, it is more expensive than CRF mouse models requiring tumor cell implantation.

Regardless of whether subcutaneous implantation, intravenous injection of CT26 cells, or orthotopic tumor transplantation is used, researchers have reported that tumor-bearing mice exhibit important fatigue behaviors, such as reduced grip strength and motor ability, decreased body weight, muscle mass and fat content, and elevated levels of inflammatory factors. Murphy *et al* (10) also found that injecting the same number of CT26 cells from different laboratories induced varying degrees of cachexia in mice, suggesting that researchers should ensure the use of cells from the same source within a single experiment to avoid excessive experimental error. Furthermore, to meet experimental objectives, it is recommended to use the minimum number of cancer cells necessary for implantation, reduce the tumor burden in mice, and perform euthanasia when tumors become excessively large, in compliance with animal ethics guidelines and to protect the welfare of laboratory animals.

Lung cancer-reduced CRF mouse models. Lewis Lung Carcinoma (LLC) is a commonly used mouse lung adenocarcinoma cell line, known for its high invasiveness and ability to simulate the progression of lung adenocarcinoma in mice. Scott *et al* (19) and Zhu *et al* (20) established lung cancer mouse models by subcutaneously implanting $5x10^5$ LLC cells in the backs of male C57BL/6J and BALB/c mice, respectively. Zhou *et al* (21) subcutaneously implanted $3x10^5$ LLC cells in the right axilla of male C57BL/6 mice, while Vichaya *et al* (22) implanted $1x10^6$ LLC cells in the axilla, successfully creating lung CRF models in mice. Although the location and number of subcutaneously implanted LLC cells varied among the aforementioned studies, all reported that tumor-bearing mice exhibited significant fatigue symptoms, such as reduced wheel running, decreased forelimb grip strength and swimming time, and elevated levels of inflammatory factors.

Apart from subcutaneous implantation of LLC cells, intraperitoneal injection has also been utilized to establish a CRF model of lung cancer. Wu *et al* (23) injected $1x10^7$ LLC cells into female ICR mice to create a fatigue model. Similarly, Kim *et al* (24) administered $0.5x10^6$ LLC cells via intraperitoneal injection in both male and female C57BL/6J mice to establish a fatigue model. Additionally, another group of mice received an injection of $0.1x10^6$ TC-1 cells (tumorigenic mouse lung epithelial cells), which also exhibited symptoms of fatigue, such as lethargy and reduced grooming behavior, although the level of fatigue was milder compared with that of the LLC group. While there are studies reporting the use of LLC cells via intrathoracic injection, these have not been applied to CRF research. Previous studies have employed varying cell numbers and implantation methods to develop mouse models of CRF, all reporting different fatigue symptoms. However, the differences in cell quantities, implantation sites and fatigue assessment methods make it challenging to compare the effects of tumor burden on the onset and severity of fatigue. Therefore, standardization of cell dosage and monitoring parameters is necessary to improve evaluation of fatigue levels in mice, and to facilitate comparison and reference across studies.

Head and neck cancer-reduced CRF mouse models. HPV16 E6/E7 and H-ras (mEER) cells are commonly used cancer cell lines closely associated with the development of head and neck cancer. Vichaya *et al* (25) established an ectopic syngeneic mouse model of HPV-related head and neck cancer using male oropharyngeal epithelial cells derived from stable expression of mEER cells. The method involved subcutaneously injecting $1x10^6$ mEER cells into the right hind leg of adult male C57BL/6J mice and $Ido1^{-/-}$ mice, resulting in reduced wheel running and fatigue symptoms in both groups. In another study, Vichaya *et al* (22) used the same method to establish a fatigue model in $Tlr4^{-/-}$ mice on a C57BL/6J background. Grossberg *et al* (26,27) reported the successful establishment of an HPV-positive head and neck cancer model

by subcutaneously implanting $1x10^6$ mEER cells into male C57BL/6J mice, while another group of mice was injected with the same number of shPTPBL/hRas cells to establish an HPV-negative head and neck cancer model, with both groups showing fatigue symptoms. Grossberg *et al* (26,27) also found that metabolic competition between skeletal muscle and tumors may represent a non-inflammatory mechanism of cancer-induced fatigue, offering new insights for its treatment. However, it is important to note that mEER cells are derived from the tonsillar epithelium of male mice, and injecting these cells into female mice may result in unsuccessful tumor implantation.

Other cancer-reduced CRF mouse models. The KPC cell line (a pancreatic cancer cell line derived from mouse pancreatic tissue with Kras deletion and p53 mutation, KrasG12D/p53R172H) is commonly used for pancreatic cancer research. Zhu *et al* (28) divided C57BL/6J mice into three groups: Two groups received either $3x10^6$ KPC cells for pancreatic orthotopic implantation or intraperitoneal injection, and the third group was subcutaneously injected with $10x10^6$ KPC cells into the shoulder joint space. Fatigue behaviors were observed as early as 6 days post-injection. Zhu *et al* (28) concluded that the orthotopic pancreatic implantation model is the most stable and reliable, while the subcutaneous model yielded inconsistent results and is not recommended. Delitto *et al* (29) also reported that, compared with intraperitoneal injection, orthotopic implantation of KPC cells led to more severe muscle atrophy and systemic inflammation.

The 4T1 cell line is a commonly used breast cancer cell line. Yang *et al* (30) successfully established a cancer-induced fatigue model by injecting $5x10^5$ 4T1 cells into the right fourth mammary fat pad of female BALB/c mice. Consistent with the results of Norden *et al* (14), Yang *et al* (30) also found that food intake and appetite were more stable in female mice compared with male mice after cancer cell inoculation, making them more suitable for cancer-induced fatigue studies.

The B16-F10 cell line is a melanoma cell line, and the ID8 cell line is a commonly used mouse ovarian cancer cell line, both of which are highly invasive. Kim *et al* (24) intraperitoneally injected $0.3x10^6$ B16-F10 and $1x10^6$ ID8 cells into two groups of C57BL/6J mice, respectively, and both groups displayed fatigue symptoms such as lethargy, reduced grooming behavior and lower hemoglobin levels, which indicate successful establishment of cancer-induced fatigue models.

Despite various cancer cell lines can be used to establish cancer-induced fatigue mouse models, most are ectopically implanted and may not fully capture the fatigue symptoms caused by organ-specific tumors. Future research should focus on cancer-induced fatigue models established by orthotopic cancer cell implantation. Detailed information on mouse models of CRF induced by tumor bearing is shown in Table I.

3. Chemotherapy-induced CRF mouse models

5-Fluorouracil (5-FU)-induced CRF mouse model. 5-FU is a commonly used antineoplastic drug that can cause severe gastrointestinal reactions and bone marrow suppression, leading to significant fatigue symptoms in patients. C57BL/6 mice are frequently used in fatigue experiments induced by intraperitoneal injection of 5-FU. Wolff *et al* (31) induced fatigue-like behavior in mice by administering 50 mg/kg 5-FU intraperitoneally for 3 consecutive days. Fatigue symptoms, including reduced body weight and spontaneous wheel running activity, were observed 1 week later. Dougherty *et al* (32) injected 60 mg/kg 5-FU intraperitoneally for 5 consecutive days, resulting in decreased body weight day 6, with voluntary wheel running activity and treadmill fatigue test confirming fatigue in the mice. Mahoney *et al* (33,34) reported in two studies that intraperitoneal injections of 5-FU at doses of 20, 40 or 60 mg/kg for 5 consecutive days led to milder fatigue and faster recovery in mice treated with lower doses, suggesting that fatigue may be dose-dependent. Hsu *et al* (35) reported that intraperitoneal injections of 60 mg/kg 5-FU for 5 consecutive days resulted in significant reductions in skeletal muscle and epididymal fat weight, as well as a marked decrease in exercise distance. Researchers have successfully established CRF mouse models using intraperitoneal injection of 5-FU. This method is simple, has a high success rate, and produces stable and reliable models. However, the appropriate 5-FU dose should be selected based on the experimental objectives to avoid excessive symptom burden and toxic reactions in the mice due to high doses of 5-FU.

Platinum-based chemotherapy-induced CRF mouse models. Cisplatin and oxaliplatin are commonly used platinum-based drugs in clinical practice, both of which can cause severe gastrointestinal reactions, bone marrow suppression and neurotoxicity, leading to significant fatigue symptoms in patients. Feather *et al* (36) established a fatigue model by administering oxaliplatin to C57BL/6J mice using both short- and long-term protocols. The short-term protocol involved administering 5 mg/kg oxaliplatin every other day for a total of four injections, with a cumulative dose of 20 mg/kg. The long-term protocol consisted of administering 2.5 mg/kg oxaliplatin 12 times over 17 days, with a cumulative dose of 30 mg/kg. The results showed that fatigue symptoms, including weight loss and reduced activity, appeared as early as the fourth day after oxaliplatin injection, with the severity of fatigue significantly correlated with the drug dose. Sakai *et al* (37) administered 3 mg/kg cisplatin intraperitoneally to C57BL/6J mice once daily for 4 consecutive days, resulting in muscle atrophy and pronounced fatigue, suggesting that fatigue may be associated with chemotherapy-induced muscle atrophy. Scott *et al* (38) and Chelette *et al* (39) administered 2.83 mg/kg cisplatin intraperitoneally to mice once daily for a total of five doses, leading to fatigue symptoms characterized by reduced wheel running activity and weight loss, but no decrease in food intake. This finding suggests that cisplatin-induced fatigue may be due to muscle fatigue rather than mental fatigue. Researchers generally use similar cumulative doses of platinum-based drugs and prefer intraperitoneal injection due to its simplicity. However, injecting these drugs may not be suitable for studying mental fatigue in mice. It is recommended that the method of fatigue induction is selected appropriately according to the research objectives.

Doxorubicin chemotherapy-induced CRF mouse models. Doxorubicin is a broad-spectrum antitumor drug with potent cytotoxicity, leading to severe bone marrow suppression and

Table I. Tumor-bearing mouse models of cancer-related fatigue.

First author, year	Mice sex and strain	Mice age	Cell line	Cell number	Site and method of cell injection	(Refs.)
Pin *et al*, 2022	Male CD2F1 mice	11 weeks	C26	$1x10^6$	Subcutaneous injection into the flank	(5)
Peters *et al*, 2011	Male CD2F1 mice	6-7 weeks	C26	$1x10^6$	Subcutaneous injection into the right inguinal flank	(6)
Shen *et al*, 2019	Male BALB/c mice	6-8 weeks	C26	$1x10^6$	Subcutaneous injection into the flank	(7)
Clark *et al*, 2015	Female CD2F1 mice	Adult	C26	$5x10^5$	Subcutaneous injection between the scapulae	(8)
Lee *et al*, 2021	BALB/c mice	6 weeks	C26	$5x10^5$	Subcutaneous injection into the right flank	(9)
Murphy *et al*, 2012	Male CD2F1 mice	12 weeks	C26	$5x10^5$	Subcutaneous injection into the dorsal side	(10)
Roberts *et al*, 2013	Male CD2F1 mice	Adult	C26	$5x10^6$	Subcutaneous injection into each flank	(12)
Song *et al*, 2024	Male BALB/c mice	6 weeks	C26	$5x10^5$	Subcutaneous injection in the dorsal region	(13)
Norden *et al*, 2015	Female CD2F1 mice	10 weeks	C26	$5x10^5$	Subcutaneous injection between the scapulae	(11,14)
Ferdaos *et al*, 2023	Male BALB/c mice	8 weeks	C26	$5x10^5$, $2x10^6$	Intravenous injection, subcutaneous injection into the right flank	(15)
Zhang *et al*, 2019	Male BALB/c mice	6-7 weeks	C26 subcutaneous tumor tissue	2x2x1 mm	Intraperitoneal orthotopic implantation	(16)
Scott *et al*, 2022	Male C57BL/6J mice	10 weeks	LLC	$5x10^5$	Subcutaneous injection into the right flank	(19)
Zhu *et al*, 2019	CD2F1 mice	4-6 weeks	LLC	$5x10^5$	Subcutaneous injection in the scapula	(20)
Zhou *et al*, 2020	Male C57BL/6J mice	6 weeks	LLC	$3x10^6$	Subcutaneous injection into the right underarm	(21)
Vichaya *et al*, 2020	C57BL6/J mice	Adult	LLC	$1x10^6$	Subcutaneous injection into the right flank	(22)
Wu *et al*, 2022	C57BL/6J mice	5 weeks	LLC	$1x10^6$	Subcutaneous injection in the right flank	(23)
Kim *et al*, 2014	C57BL/6J mice	6 weeks	LLC, TC-1, B16-F10, ID8	$0.5x10^6$, $0.1x10^6$, $0.3x10^6$, $1x10^6$	Intraperitoneal injection	(24)
Vichaya *et al*, 2019	Male C57BL/6J mice, Ido1-/- mice	Adult	mEER, luciferase-tagged mEER	$1x10^5$, $1x10^6$	Intramuscular injection into the right hind leg, subcutaneous injection into the right flank	(25)
Grossberg *et al*, 2020	Male C57BL/6J mice	10-13 weeks	mEER	$1x10^6$	Subcutaneous injection into the right flank	(26)
Grossberg *et al*, 2018	Male C57BL/6J	10-13 weeks	mEER, LLC, ID8, IG10, shPTPBL/hRas	$1.1x10^6$, $5x10^5$, $1x10^6$, $1x10^6$, $1x10^6$	Subcutaneous injection into the right flank	(27)
Zhu *et al*, 2019	C57BL/6J mice	Adult	KPC	$3x10^6$, $3x10^6$, $10x10^6$	Orthotopic implantation, intraperitoneal injection, subcutaneous injection into interscapular space	(28)
Delitto *et al*, 2017	Female NSG mice	8 weeks	PANC-1, L3.6pl	$1x10^6$	Orthotopic injections into the pancreas, subcutaneous injection	(29)
Yang *et al*, 2021	Female BALB/c mice	10 weeks	4T1	$5x10^5$	Subcutaneous injection into the right fourth mammary fat pad	(30)

LLC, Lewis lung carcinoma; mEER, HPV16 E6/E7 and H-ras; sh, small hairpin.

cardiotoxicity. Gilliam *et al* (40) administered doxorubicin to C57BL/6 mice through both intravenous and intraperitoneal routes at a dose of 20 mg/kg (equivalent to the clinical dose for patients) for 3 consecutive days. The study found that doxorubicin caused hind limb muscle impairment in mice, with both administration routes leading to respiratory muscle weakness, although intraperitoneal injection resulted in more severe diaphragm weakness. Mackay *et al* (41) conducted an experiment with a single intraperitoneal injection of 15 mg/kg doxorubicin and observed no significant muscle strength decline after 3 days, possibly due to the short observation period, during which skeletal muscle dysfunction had not yet fully developed. Zombeck *et al* (42) administered doxorubicin to female Hsd-ICR (CD1) mice at a dose of 2.5 mg/kg per day for 6 days. By the second week of treatment, the mice exhibited weight loss and reduced afternoon wheel running activity, similar to the afternoon fatigue exacerbation observed in patients with cancer. Wang *et al* (43) created a circadian rhythm disorder environment by altering light exposure and administered doxorubicin intraperitoneally at 3.75 mg/kg on days 1, 2, 9 and 10. The mice showed weight loss, decreased wheel running activity and extended rest periods, with fatigue symptoms associated with immunosuppression and circadian rhythm disturbances. Wang *et al* (44) further reported that, when comparing doxorubicin administered at 3.75 mg/kg intraperitoneally with 3.75 mg/kg daunorubicin and 11.25 mg/kg etoposide using the same method, the doxorubicin group exhibited more severe fatigue symptoms. Although numerous researchers have used doxorubicin to establish CRF mouse models, due to the severe cytotoxicity of doxorubicin and the more pronounced toxic reactions observed with intraperitoneal injection, caution must be exercised regarding the dosage and duration of experiments following doxorubicin administration. Some researchers have reported that a 20 mg/kg intraperitoneal injection of doxorubicin can result in 60-70% mortality in wild-type mice within 1 week (45,46).

Taxane drugs or cyclophosphamide-induced CRF mouse models. Taxane drugs are commonly used in clinical chemotherapy and can cause bone marrow suppression, neurotoxicity and allergic reactions. Ray *et al* (47) divided female BALB/c mice into two groups, and administered paclitaxel and albumin-bound paclitaxel (Abraxane) via the retro-orbital sinus vein at a dose of 10 mg/kg once daily for 5 consecutive days. The results showed that, while both groups of mice exhibited reduced activity indicative of fatigue, the paclitaxel group showed muscle damage and more severe fatigue. Loman *et al* (48) administered paclitaxel at 30 mg/kg (80% of the intravenous dose used in patients) to female BALB/c mice via tail vein injection every other day for a total of 6 doses. The study found that the mice's physical activity significantly decreased. Grant *et al* (49) used the same method and dosage to administer paclitaxel to female C57BL/6 mice, and observed that paclitaxel-induced fatigue in mice gradually recovered over time. Chaillou *et al* (50) administered a single dose of 20 mg/kg docetaxel and weekly injections of 20 mg/kg for 3 weeks via tail vein injection to female FVB/NRj mice. The results showed no weight difference between the two groups, but muscle mass significantly decreased in mice that underwent multiple chemotherapy sessions compared with that in mice that received a single dose. All the aforementioned researchers used intravenous injection of taxane drugs to establish fatigue models in mice. Compared with other taxane drugs, paclitaxel-induced fatigue is more severe. Furthermore, taxane-induced fatigue is generally milder and more easily recoverable compared with doxorubicin-induced fatigue, suggesting that researchers should select appropriate taxane drugs to establish CRF mouse models based on their research objectives.

Cyclophosphamide is also a commonly used clinical chemotherapy drug, which can cause bone marrow suppression and acute skeletal muscle effects. Crouch *et al* (51) administered a single tail vein injection of 300 mg/kg cyclophosphamide to female C57BL/6 mice, resulting in reduced activity indicative of fatigue. A total of 6 weeks after administration, treadmill running time was still significantly reduced. Our systematic literature review revealed that studies on inducing fatigue in mice using cyclophosphamide alone are relatively scarce.

Combination chemotherapy-induced CRF mouse models. Combination chemotherapy regimens are the most commonly used treatments for patients with cancer, and inducing fatigue in mice with combination chemotherapy may improve simulation of clinical scenarios compared with single-agent chemotherapy. Smith *et al* (52) used the CAF regimen (cyclophosphamide 167 mg/kg, doxorubicin 4 mg/kg and 5-fluorouracil 167 mg/kg) administered via intraperitoneal injection. The method involved intraperitoneal injection of cyclophosphamide and doxorubicin, followed by intraperitoneal injection of 5-FU 1 h later, for a total of four treatments, with each chemotherapy session spaced 20 days apart. The mice showed significant weight loss and a marked reduction in voluntary wheel running activity, with pronounced fatigue symptoms. Weymann *et al* (53) also used the CAF regimen to establish a mouse model of combination chemotherapy-induced fatigue, finding that this regimen induced fatigue/somnolence and decreased voluntary motor activity in mice. Wong *et al* (54) administered the CA regimen (cyclophosphamide 167 mg/kg and doxorubicin 4 mg/kg) via intraperitoneal injection to C57BL/6 mice every 2 weeks for a total of four treatments. The mice exhibited weight loss and reduced voluntary wheel running time, but their weight returned to baseline levels after the treatment ended.

FOLFOX (5-fluorouracil, leucovorin and oxaliplatin) is also a commonly used chemotherapy regimen, known to cause fatigue, weakness and skeletal muscle dysfunction. Halle *et al* (55,56) reported in two studies that intraperitoneal injection of 30 mg/kg 5-FU, 6 mg/kg oxaliplatin and 90 mg/kg leucovorin was administered to male C57BL/6 mice every 2 weeks for a total of four treatments. The results showed a decrease in running time and forelimb grip strength in mice, but no weight loss was observed. Barreto *et al* (57) also reported a study where male CD2F1 mice were divided into FOLFOX and FOLFIRI (5-FU, leucovorin, irinotecan) groups, and underwent five cycles of chemotherapy. Both groups showed quadriceps atrophy and muscle weakness, but only the FOLFIRI group exhibited progressive weight loss. The FOLFOX regimen was shown to induce fatigue symptoms in mice, with minimal impact on body weight, suggesting that studies focusing primarily on weight changes should

carefully consider the FOLFOX chemotherapy regimen as an intervention factor.

Although both single-agent and combination chemotherapy can induce fatigue symptoms in mice and successfully establish chemotherapy-related CRF models, these experiments were conducted without tumor burden and therefore only reflect the effects of chemotherapy drugs on fatigue. Additionally, the drug dosages and injection routes used by different researchers vary. It is recommended that researchers, based on their study objectives and considering animal welfare, select appropriate methods and routes to establish CRF mouse models for research purposes. Datailed data on mouse models of CRF induced by chemotherapy are shown in Table II.

4. Tumor-bearing mice with chemoradiotherapy-induced CRF mouse models

Colon cancer cell-bearing mice with chemotherapy-induced CRF models. Compared with chemotherapy-induced fatigue in healthy mice, chemotherapy in tumor-bearing mice may improve simulation of the CRF symptoms observed in patients with cancer. Abulizi *et al* (58) implanted $2.5x10^5$ CT26 cells subcutaneously into female BALB/c mice to establish tumor-bearing mice. A total of 8 days later, 30 mg/kg of 5-FU was administered intraperitoneally every other day for a total of eight injections. Compared with the tumor-bearing mice without chemotherapy, the 5-FU chemotherapy group exhibited significantly increased immobility time in the forced swim test, indicating more severe fatigue. Furthermore, muscle histology showed disordered and ruptured skeletal muscle fibers with shortened sarcomere length. Park *et al* (59) subcutaneously inoculated $5x10^6$ HT-29 cells (human-derived colon cancer cells) into immunodeficient female BALB/c-nu/nu mice. A total of 14 days later, 30 mg/kg of 5-FU was administered intraperitoneally 3 times a week for a total of 12 injections. The results similarly demonstrated that the 5-FU chemotherapy group exhibited significantly lower performance in both the running wheel activity and forced swim test compared with the tumor-bearing group without chemotherapy. Additionally, the 5-FU chemotherapy group had lower white and red blood cell counts. Compared with tumor-bearing mice without chemotherapy, 5-FU chemotherapy exacerbated fatigue in CT26 tumor-bearing mice. This model of CRF induced by 5-FU in colon cancer-bearing mice better replicates the clinical situation of chemotherapy in patients with colon cancer, and is more suitable for exploring the mechanisms of CRF and for developing new drugs.

In a study on alleviating fatigue induced by weighted swimming in rats, Wang *et al* (60) employed gas chromatography-mass spectrometry (MS)-based metabolomics and found significant metabolic disturbances in fatigued rats. Specifically, lipid peroxidation products, such as malondialdehyde, were markedly elevated, while levels of glutathione and superoxide dismutase were decreased, indicating an imbalance in skeletal muscle energy metabolism. In a study on postoperative fatigue syndrome, Lu *et al* (61) used ultra-high performance liquid chromatography-quadrupole time-of-flight MS-based metabolomics and identified hippocampal metabolic fluctuations in postoperative fatigued rats. The authors identified 15 hippocampal metabolites that could serve as potential drug targets, highlighting metabolomics as an important tool in the study of fatigue mechanisms. Future research could further explore the application of metabolomics in understanding the mechanisms of fatigue, providing a more diverse range of tools for fatigue mechanism research.

Lung cancer cell-bearing mice with chemotherapy-induced CRF models. Lung cancer-bearing mice treated with chemotherapy are also commonly used to induce CRF. Wu *et al* (23) subcutaneously implanted $1x10^6$ LLC cells into C57BL/6 mice. When the tumor size reached 80-100 mm^3, cisplatin was administered intraperitoneally at a dose of 5 mg/kg once a week for 3 weeks. Compared with tumor-bearing mice without chemotherapy, the cisplatin-treated group showed significantly reduced swimming time and distance, indicating more pronounced fatigue. Yoshizawa *et al* (62) established a similar tumor-bearing model by subcutaneously implanting cancer cells and, 8 days later, administering cisplatin intraperitoneally at 10 mg/kg for 4 consecutive days. The cisplatin-treated mice exhibited significantly reduced running time on the treadmill test and experienced weight loss compared with the tumor-bearing group without chemotherapy; however, their weight returned to baseline by day 16. Ouyang *et al* (63) subcutaneously implanted $5x10^6$ A549 human lung adenocarcinoma cells into male BALB/c-nu nude mice. When the tumor size reached 80-100 mm^3, cisplatin was administered intraperitoneally at 10 mg/kg every other day for a total of 10 injections. Compared with the tumor-bearing group without chemotherapy, the cisplatin-treated group experienced earlier onset of fatigue and higher levels of inflammatory cytokines. Although the cisplatin dosage and treatment duration varied across studies, the results consistently showed that cisplatin exacerbated and accelerated the onset of fatigue in tumor-bearing mice, with its effects on body weight gradually reversing over time.

Wood *et al* (64) subcutaneously implanted $5x10^5$ LLC cells into female C57BL/6 mice to establish a tumor-bearing mouse model. A total of 8 days after cancer cell implantation, the mice were administered etoposide intraperitoneally at a dose of 50 mg/kg every 3 days for a total of five doses. Compared with the tumor-bearing group without chemotherapy, the etoposide-treated group showed no significant differences in voluntary wheel-running activity or body weight. However, the chemotherapy group had lower hemoglobin levels and higher serum IL-6 levels, suggesting that etoposide has a limited impact on fatigue in tumor-bearing mice. Therefore, caution is advised when selecting this drug for modeling CRF.

Although several researchers have studied fatigue models in lung cancer-bearing mice treated with chemotherapy, there are considerable variations in the number of cancer cells implanted, as well as in the dosage and duration of the chemotherapy regimens used. These differences make it difficult to compare the extent of fatigue across studies or to determine the specific impact of chemotherapy dosage on fatigue. Future research should focus on exploring the effects of varying chemotherapy doses on fatigue in lung cancer-bearing mice. Currently, gene editing technologies have been applied in fatigue mouse models, but their use has largely been limited to the establishment of CRF models by implanting human-derived tumor cells into nude mice. Future

Table II. Chemotherapy-induced cancer-related fatigue mouse models.

First author, year	Mice sex and strain	Mice age	Chemotherapy drugs and dosage	Chemotherapy duration	Administration method	(Refs.)
Wolff *et al*, 2022	BNDF mice	7-18 weeks	5-FU 50 mg/kg	3 consecutive days	Intraperitoneal injection	(31)
Mahoney *et al*, 2013	C57BL/6 mice, MCP-1-/- mice	6 weeks	5-FU 20 mg/kg, 5-FU 40 mg/kg, 5-FU 60 mg/kg	5 consecutive days	Intraperitoneal injection	(33)
Hsu *et al*, 2020	Male C57BL/6 mice	8-9 weeks	5-FU 60 mg/kg	5 consecutive days	Intraperitoneal injection	(35)
Feather *et al*, 2018	Male C57BL/6 mice	8 weeks	Oxaliplatin 5 mg/kg, oxaliplatin 2.5 mg/kg	Once every other day, total of four doses, 3 consecutive days followed by a 3-day break for a total of nine doses	Intraperitoneal injection	(36)
Sakai *et al*, 2014	Male C57BL/6J mice	8-9 weeks	Cisplatin 1 mg/kg, cisplatin 3 mg/kg	4 consecutive days	Intraperitoneal injection	(37)
Scott *et al*, 2023	C57BL/6J mice	8 weeks	Cisplatin 2.83 mg/kg	5 consecutive days	Intraperitoneal injection	(38)
Chelette *et al*, 2023	C57BL/6J mice	10-12 weeks	Cisplatin 2.83 mg/kg	5 consecutive days	Intraperitoneal injection	(39)
Gilliam *et al*, 2011	Male C57BL/6 mice	8 weeks	Doxorubicin 20 mg/kg	One single dose	Intravenous administration, intraperitoneal injection	(40)
Mackay *et al*, 2021	C57BL/6 mice	5 weeks	Doxorubicin 15 mg/kg	One single dose	Intraperitoneal injection	(41)
Zombeck *et al*, 2013	Female Hsd-ICR mice	7 weeks	Doxorubicin 2.5 mg/kg	3 consecutive days, repeated after 1 week for a total of six doses	Intraperitoneal injection	(42)
Wang *et al*, 2024	Male C57BL/6J mice	10 weeks	Doxorubicin 3.75 mg/kg, aclarubicin 3.75 mg/kg, etoposide 11.25 mg/kg	Days 1, 2, 9 and 10	Intraperitoneal injection	(43)
Ray *et al*, 2011	Female BALB/c mice	Adult	Paclitaxel 10 mg/kg, Abraxane 10 mg/kg	5 consecutive days	Retro-orbital sinus vein injection	(47)
Loman *et al*, 2019	Female BALB/c mice	7-8 weeks	Paclitaxel 30 mg/kg	Once every other day for a total of six doses	Caudal vein injection	(48)
Grant *et al*, 2023	Female C57BL/6 mice	12 weeks	Paclitaxel 30 mg/kg	Once every other day for a total of six doses	Tail vein injection	(49)
Chaillou *et al*, 2017	Female mice on FVB/NRj strain background	8-11 weeks	Docetaxel 20 mg/kg	One single dose, days 1 and 21	Tail vein injection	(50)
Crouch *et al*, 2017	Female C57BL/6J mice	16 weeks	Cyclophosphamide 300 mg/kg	One single dose	Tail vein injection	(51)

Table II. Continued.

First author, year	Mice sex and strain	Mice age	Chemotherapy drugs and dosage	Chemotherapy duration	Administration method	(Refs.)
Smith *et al*, 2014	Female mice lacking TNFR1-/- and/or IL-1R1-/-	Adult	CAF regimen (cyclophosphamide 167 mg/kg, doxorubicin 4 mg/kg, 5-FU 167 mg/kg)	Four doses of CAF at 10-20 days intervals	Intraperitoneal injection	(52)
Weymann *et al*, 2014	Female C57BL/6J mice	8-12 weeks	CAF regimen (cyclophosphamide 167 mg/kg, doxorubicin 4 mg/kg, 5-FU 167 mg/kg)	Four cycles at 3-week intervals	Intraperitoneal injection	(53)
Wong *et al*, 2018	Female C57BL/6 mice	10-12 weeks	CA regimen (cyclophosphamide 167 mg/kg, doxorubicin 4 mg/kg)	One single dose	Intraperitoneal injection	(54)
Halle *et al*, 2023	Male C57BL6/J mice	10 weeks	FOLFOX regimen (5-FU 30 mg/kg oxaliplatin, 6 mg/kg, leucovorin 90 mg/kg)	Four cycles at 2-week intervals	Intraperitoneal injection	(56)

5-FU, 5-fluorouracil.

research should further explore the application of gene editing techniques to knock out or overexpress genes associated with CRF and observe their effects on fatigue symptoms. This approach could provide potential therapeutic targets for subsequent targeted treatments of CRF.

Breast cancer cell tumor-bearing and chemotherapy-induced CRF mouse models. Lv *et al* (65) subcutaneously injected $2x10^5$ 4T1 cells into female BALB/c mice. When the tumor volume reached ~0.5 cm^3, cyclophosphamide was administered intraperitoneally at a dose of 100 mg/kg once daily for 3 consecutive days. Mice that were both tumor bearing and received chemotherapy exhibited symptoms of fatigue, including weight loss, reduced appetite, lethargy, slowed movement, prolonged immobility in the forced swim test and decreased scores in both horizontal and vertical movement in the open field test. Additionally, there was a significant increase in inflammatory cytokines in both the intestine and hippocampus.

Ouyang *et al* (63) used a similar approach, injecting $1x10^5$ 4T1 cells into mice. When the tumor volume reached 50-100 mm^3, paclitaxel was administered intraperitoneally at a dose of 10 mg/kg every other day for a total of 10 doses. Compared with the group with only tumors, the combined tumor-bearing and chemotherapy group showed a more pronounced reduction in weight-loaded swimming time, indicating more severe fatigue, along with higher serum levels of inflammatory cytokines. Although the number of 4T1 cells implanted subcutaneously differed by a factor of two between these studies, both showed a high tumor formation rate. Further research is needed to determine the optimal number of tumor cells that can meet research requirements without causing excessive tumor growth to ensure animal welfare.

Currently, mouse fatigue in CRF studies is assessed using subjective indicators such as anorexia, drowsiness and sluggish movement, which may lead to inconsistencies in judgment standards among different researchers. Rafaih and Ari (66) applied artificial intelligence (AI)-driven technology through machine learning algorithms, wearable devices and real-time feedback systems to analyze the physiological, behavioral and environmental data of surgeons for comprehensive fatigue monitoring. However, there is limited research on using AI technology to monitor mouse fatigue behavior. Future studies could explore the use of AI technology to more accurately and comprehensively assess and monitor mouse fatigue.

Head and neck tumor cell-bearing with chemoradiotherapy-induced CRF mouse models. Chemoradiotherapy is a common treatment strategy for head and neck cancer, and patients may develop fatigue within the first few days of combined treatment (67). Vichaya *et al* (68,69) injected $1x10^5$ mEER cells into the right hind limb of male C57BL/6 mice. On the seventh day post-cell implantation, cisplatin was administered intraperitoneally at a dose of 5.28 mg/kg, combined with 8 Gy of localized leg radiation therapy once a week for 4 weeks. The mice exhibited burrowing deficits, weight loss and increased inflammatory cytokines in liver and tumor tissues. Vichaya *et al* (70) also reported implanting mEERL95 cells into C57BL/6J mice, noting that this cell line is applicable to both male and female mice. The study found

Table III. Chemotherapy and/or radiotherapy-induced cancer-related fatigue mouse models with tumor burden.

First author/s, year	Mice sex and strain	Mice age	Cell line and number	Cell injection site and method	Chemotherapy drugs and/or dosage	Chemotherapy and/or radiotherapy duration	(Refs.)
Wu *et al*, 2022	C57BL/6J mice	5 weeks	LLC $1x10^6$	Subcutaneous injection in the right flank	Cisplatin 5 mg, intraperitoneal injection	Once per week over a period of 3 weeks	(23)
Wang *et al*, 2024	Swiss mice	Adult	H22 $1x10^7$	Subcutaneous injection into the right flank	Cisplatin 10 mg/kg, intraperitoneal injection	Once every other day for a total of seven doses	(43)
Abulizi *et al*, 2021	Female BALB/c mice	6-7 weeks	C26 $2.5x10^5$	Subcutaneous injection over the scapula	5-FU 30 mg/kg, intraperitoneal injection	Every other day for a total of eight doses	(58)
Park *et al*, 2015	Female BALB/c-nu/nu mice	5 weeks	HT-29 $5x10^6$	Subcutaneous injection in the right flank	5-FU 30 mg/kg, intraperitoneal injection	3 consecutive days per week over a period of 4 weeks	(59)
Yoshizawa *et al*, 2021	Male C57BL/6N mice	7 weeks	LLC $1x10^6$	Subcutaneous injection in the right flank	Cisplatin 10 mg/kg, intraperitoneal injection	One single dose	(62)
Ouyang *et al*, 2016	Male BALB/c-nu mice	5-6 weeks	A549 $5x10^6$	Subcutaneous injection in the right flank	Cisplatin 10 mg/kg, intraperitoneal injection	Once every 2 days for a total of 10 doses	(63)
Ouyang *et al*, 2016	Female BALB/c mice	7 weeks	4T1 $1x10^5$	Subcutaneous injection into the fourth breast pad area	Paclitaxel 10 mg/kg, intraperitoneal injection	Once every other day for a total of 10 doses	(63)
Wood, *et al*, 2006	Female C57BL/6J mice	8 weeks	LLC $5x10^5$	Subcutaneous injection in the scapular region	VP-16 50 mg/kg, intraperitoneal injection	Once every 3 days for a total of five doses	(64)
Lv *et al*, 2022	Female BALB/c mice	6-8 weeks	4T1 $2x10^5$	Subcutaneous injection into the fourth breast pad area	Cyclophosphamide 100 mg/kg, intraperitoneal injection	Once daily for 3 consecutive days	(65)
Vichaya *et al*, 2021	Male C57BL/6J mice	Adult	mEER $1x10^6$	Intramuscular injection into the right hind leg	Cisplatin 5.28 mg/kg, intraperitoneal injection and local radiation 8 Gy	Once per week over a period of 4 weeks	(70)
Phan *et al*, 2024	C57BL/6J mice	8 weeks	mEER $1x10^6$, LLC $5x10^5$	Intramuscular injection into the right hind leg, subcutaneous injection in the right flank	Cisplatin 5.28 mg/kg, intraperitoneal injection and local radiation 8 Gy	Once per week over a period of 4 weeks	(71)

LLC, Lewis lung carcinoma; mEER, HPV16 E6/E7 and H-ras; 5-FU, 5-fluorouracil.

Table IV. Peripheral irradiation-induced cancer-related fatigue mouse models.

First author/s, year	Mice sex and strain	Mice age	Peripheral irradiation dosage and area	Duration of peripheral irradiation	(Refs.)
Renner *et al*, 2016	Male C57BL/6 mice	5 weeks	8 Gy external lower abdominal irradiation	Once daily for a total of 3 days	(78)
McDonald *et al*, 2016	Male C57BL/6 mice	10-12 weeks	2.84 Gy external pelvic irradiation	5 consecutive days per week for a total of 24 times	(79)
Wood *et al*, 2008	Male C57BL/6 mice	Adult	2.84 Gy external pelvic irradiation	5 consecutive days per week for a total of 24 times	(80)
Huang *et al*, 2022	Male BALB/c mice	4 weeks	2 Gy internal left adrenal gland irradiation	One single time	(81)

that female mice had smaller tumors and exhibited fatigue behaviors later and less frequently than males, suggesting the importance of considering sex differences in the fatigue levels of head and neck tumor-bearing mice.

Phan *et al* (71) divided mice into two groups: One was injected with mEER cells and the other one with LLC cells. Post-chemoradiotherapy, both groups experienced weight loss and reduced running wheel activity, with fatigue symptoms worsening as the number of chemoradiotherapy sessions increased. These results indicate that chemoradiotherapy exacerbates fatigue in a dose-dependent manner, highlighting the need to tailor the dose and regimen of chemotherapeutic agents according to the research objectives.

Other tumor cell-bearing with chemotherapy-induced CRF mouse models. Wang *et al* (72) injected $1x10^7$ H22 hepatocellular carcinoma cells subcutaneously into Swiss mice. When the subcutaneous tumor reached 80-100 mm^3, cisplatin was administered intraperitoneally at a dose of 10 mg/kg every other day for a total of seven doses. Compared with tumor-bearing mice without chemotherapy, the chemotherapy group exhibited more significant reductions in body weight, load-bearing swim time and activity levels, along with higher serum BUN and IL-1β levels.

Zhang *et al* (73) divided male C57BL/6 mice into two groups. One group was subcutaneously implanted with $5x10^5$ YUMMER1.7 malignant melanoma cells, and on day 13 post-implantation, the mice received intraperitoneal injections of anti-mouse PD-1 antibody (BioXCell, InVivoMab series, clone RMP1-14) at 10 mg/kg three times a week. The other group was subcutaneously implanted with $1x10^6$ MC38 colon cancer cells, and on day 12 post-implantation, the mice received intraperitoneal injections of 5-FU at 100 mg/kg for chemotherapy three times a week. The study showed that anti-PD-1 immunotherapy impaired the motivation for activity, leading to reduced forced swim time, while chemotherapy impaired muscle function, resulting in decreased forelimb grip strength and a reduction in maximum treadmill speed. These results suggest that, although both immunotherapy and chemotherapy can induce fatigue symptoms in mice, the underlying mechanisms differ, leading to distinct fatigue behaviors. Given the limited research on immunotherapy-related fatigue, further studies are needed to elucidate the mechanisms and behavioral manifestations of fatigue induced by immunotherapy. Detailed data on mouse models of CRF induced by chemotherapy and/or radiotherapy with tumor burden are shown in Table III.

Chen *et al* (74) administered anti-PD-1 antibody (12.5 μg/g every 5 days for 6 injections) and anti-PD-L1 antibody (10 μg/g once a week for 6 weeks) to 6-8-week-old C57BL/6 mice, resulting in significant cardiac toxicity symptoms, including myocarditis. Cao *et al* (75) found that anti-PD-1 treatment led to DNA and mitochondrial damage in cardiac macrophages, and could induce macrophage polarization towards a pro-inflammatory phenotype. Other researchers also observed that anti-PD-1 treatment caused heart failure, left ventricular dilation and enhanced thymic inflammation signaling in mice. Inflammation in the myocardium and thymus may further exacerbate fatigue symptoms in mice. Currently, animal models related to immune checkpoint inhibitor therapy mainly

focus on myocarditis models, with fewer studies investigating the induction of fatigue by immune checkpoint inhibitors in mice. Further attention is needed to explore the mechanisms underlying fatigue induced by immunotherapy.

5. Peripheral irradiation-induced CRF mouse models

Peripheral irradiation may exacerbate fatigue in patients with cancer. Wolff *et al* (76) performed localized irradiation on the lower abdominal/pelvic region of anesthetized male C57BL/6 mice with a dose of 8 Gy for 3 consecutive days. The results indicated reduced activity and the appearance of fatigue symptoms in the mice, accompanied by weight loss. Wolff *et al* (31,77) also reported that voluntary running wheel distance was minimized on day 3 post-radiation therapy, with a concomitant reduction in brain-derived neurotrophic factor (BDNF) levels in pelvic-irradiated mice. The decrease in BDNF may be causally linked to fatigue. Renner *et al* (78) used a similar peripheral irradiation method to establish a murine model of fatigue. Their findings showed that the fatigue trajectory in mice closely mirrored that observed in clinical patients, with reduced running distance and speed on the voluntary running wheel in the afternoon or evening, indicating exacerbated fatigue during this period. This animal model realistically simulates clinical conditions and offers an suitable model for studying fatigue induced by peripheral irradiation.

McDonald *et al* (79) conducted external beam radiation therapy (EBRT) on male C57BL/6J mice, delivering a dose of 2.84 Gy five times a week for a total of 24 sessions. This treatment regimen mimicked the radiotherapy protocols used in clinical patients with prostate cancer. The results indicated that the mice subjected to EBRT exhibited symptoms of fatigue, including reduced voluntary wheel running time and distance, weight loss, decreased food intake, and elevated levels of inflammatory markers in blood and tissues. Similarly, Wood *et al* (80) employed an analogous pelvic irradiation method, supporting these findings. Huang *et al* (81) anesthetized male BALB/c mice and surgically exposed the left adrenal gland, administering a radiation dose of 2 Gy. The results demonstrated that irradiated mice experienced decreased swimming endurance and significant reductions in nighttime cortisol levels. The models of fatigue induced by localized irradiation in healthy mice established by the aforementioned researchers may not adequately reflect clinical scenarios. Additionally, the radiation doses, treatment frequencies and targeted sites varied among studies. Future research should focus on fatigue induced by localized irradiation in tumor-bearing mice, as this situation aligns more closely with clinical conditions. Detailed information on mouse models of CRF induced by peripheral irradiation is shown in Table IV.

6. Conclusions

Despite the development of fatigue mouse models through various methods such as tumor cell implantation, chemotherapy and radiation therapy, and subsequent behavioral and biological studies, the molecular mechanisms underlying CRF remain insufficiently understood. Future research could explore the integration of genomic, transcriptomic and metabolomic data through multi-omics analysis. By performing data correlation analyses at a holistic level, researchers may identify fatigue-related biomarkers and potential molecular targets associated with CRF. These findings could be validated using gene editing techniques and applied to the early prediction and monitoring of CRF, thereby providing a theoretical foundation for drug development.

Despite the development of CRF mouse models through various methods such as tumor cell implantation, chemotherapy and radiation therapy, these models often fail to accurately replicate the full spectrum of fatigue symptoms experienced by patients with cancer. Numerous models rely on ectopic tumor implantation, which does not mimic the systemic effects of orthotopic tumors. Additionally, current CRF mouse models assess fatigue symptoms in the absence of comorbidities and without considering the psychological stress responses associated with cancer and its treatments, such as anxiety, depression and pain. This limitation hinders the simulation of the psychological factors that significantly contribute to fatigue in patients with cancer. Furthermore, there is a lack of consensus regarding the assessment methods for fatigue in mice. Existing evaluations predominantly focus on physical fatigue, with minimal attention given to emotional and cognitive dimensions. This narrow focus may impede a comprehensive understanding of CRF mechanisms. Future research should aim to develop more clinically relevant mouse models by integrating cancer-induced effects with psychological stressors, such as isolation-induced anxiety or chronic social defeat-induced depression. Employing multidimensional assessment tools, including biomarkers such as inflammatory cytokines and metabolites, physiological evaluations, behavioral assays and cognitive tests (Morris water maze) can facilitate a holistic evaluation of fatigue across physical, emotional and cognitive domains. This approach will enhance the current understanding of CRF mechanisms and inform the development of targeted therapeutic strategies.

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Authors' contributions

CX designed the study and reviewed the manuscript. HG was responsible for the literature review and drafted the initial manuscript. LC conducted the literature search and acquired, analyzed and interpreted the data. All authors read and approved the final version of the manuscript. Data authentication is not applicable.

Ethics approval and consent to participate

Not applicable.

Patient consent for publication

Not applicable.

Competing interests

The authors declare that they have no competing interests.

References

1. Goedendorp MM, Gielissen MFM, Verhagen CAH, Peters MEJW and Bleijenberg G: Severe fatigue and related factors in cancer patients before the initiation of treatment. Br J Cancer 99: 1408-1414, 2008.
2. Bi Y, Liu X, Liu Y, Wang M, Shan Y, Yin Y, Meng X, Sun F, Li H and Li Z: Molecular and biochemical investigations of the anti-fatigue effects of tea polyphenols and fruit extracts of Lycium ruthenicum Murr. on mice with exercise-induced fatigue. Front Mol Biosci 10: 1223411, 2023.
3. Al Maqbali M, Al Sinani M, Al Naamani Z, Al Badi K and Tanash MI: Prevalence of fatigue in patients with cancer: A systematic review and meta-analysis. J Pain Symptom Manage 61: 167-189.e14, 2021.
4. Weis J: Cancer-related fatigue: Prevalence, assessment and treatment strategies. Expert Rev Pharmacoecon Outcomes Res 11: 441-446, 2011.
5. Pin F, Huot JR and Bonetto A: The mitochondria-targeting agent MitoQ improves muscle atrophy, weakness and oxidative metabolism in C26 tumor-bearing mice. Front Cell Dev Biol 10: 861622, 2022.
6. Peters SJ, van Helvoort A, Kegler D, Argilès JM, Luiking YC, Laviano A, van Bergenhenegouwen J, Deutz NE, Haagsman HP, Gorselink M and van Norren K: Dose-dependent effects of leucine supplementation on preservation of muscle mass in cancer cachectic mice. Oncol Rep 26: 247-254, 2011.
7. Shen Q, Miao CX, Zhang WL, Li YW, Chen QQ, Li XX, Liu X and Zhang XW: SiBaoChongCao exhibited anti-fatigue activities and ameliorated cancer cachexia in mice. RSC Adv 9: 17440-17456, 2019.
8. Clark YY, Wold LE, Szalacha LA and McCarthy DO: Ubiquinol reduces muscle wasting but not fatigue in tumor-bearing mice. Biol Res Nurs 17: 321-329, 2015.
9. Lee SB, Lee JS, Moon SO, Lee HD, Yoon YS and Son CG: A standardized herbal combination of Astragalus membranaceus and Paeonia japonica, protects against muscle atrophy in a C26 colon cancer cachexia mouse model. J Ethnopharmacol 267: 113470, 2021.
10. Murphy KT, Chee A, Trieu J, Naim T and Lynch GS: Importance of functional and metabolic impairments in the characterization of the C-26 murine model of cancer cachexia. Dis Model Mech 5: 533-545, 2012.
11. Norden DM, Bicer S, Clark Y, Jing R, Henry CJ, Wold LE, Reiser PJ, Godbout JP andf McCarthy DO: Tumor growth increases neuroinflammation, fatigue and depressive-like behavior prior to alterations in muscle function. Brain Behav Immun 43: 76-85, 2015.
12. Roberts BM, Frye GS, Ahn B, Ferreira LF and Judge AR: Cancer cachexia decreases specific force and accelerates fatigue in limb muscle. Biochem Biophys Res Commun 435: 488-492, 2013.
13. Song G, Park WY, Jiao W, Park JY, Jung SJ, Ma S, Lee J, Lee KY, Choe SK, Park J, *et al*: Moderating AKT signaling with baicalein protects against weight loss by preventing muscle atrophy in a cachexia model caused by CT26 colon cancer. Biochim Biophys Acta Mol Cell Res 1871: 119670, 2024.
14. Norden DM, Devine R, Bicer S, Jing R, Reiser PJ, Wold LE, Godbout JP and McCarthy DO: Fluoxetine prevents the development of depressive-like behavior in a mouse model of cancer related fatigue. Physiol Behav 140: 230-235, 2015.
15. Ferdaos N, Harada A, Masuda E, Kasai S, Horaguchi T and Yoshizawa K: Caffeine-supplemented diet prevents fatigue-like behavior in tumor-bearing mice. Nutr Cancer 75: 1005-1013, 2023.
16. Zhang Y, Li Z, Zhao Z, Kuai W, Wei C, Lv J, Zhi J and Jia Y: Effect of the Chinese medicine YangZheng XiaoJi on reducing fatigue in mice with orthotopic transplantation of colon cancer. Evid Based Complement Alternat Med 2019: 3870812, 2019.
17. VanderVeen BN, Hardee JP, Fix DK and Carson JA: Skeletal muscle function during the progression of cancer cachexia in the male $Apc^{Min/+}$ mouse. J Appl Physiol (1985) 124: 684-695, 2018.
18. Hetzler KL, Hardee JP, Puppa MJ, Narsale AA, Sato S, Davis JM and Carson JA: Sex differences in the relationship of IL-6 signaling to cancer cachexia progression. Biochim Biophys Acta 1852: 816-825, 2015.
19. Scott K, Phan TT, West AP, Taniguchi CM and Dantzer R: Neutralizing interleukin-6 in tumor-bearing mice does not abrogate behavioral fatigue induced by Lewis lung carcinoma. Behav Brain Res 417: 113607, 2022.
20. Zhu G, Zhang B, Jiang F, Zhao L and Liu F: ShenQi FuZheng Injection ameliorates fatigue-like behavior in mouse models of cancer-related fatigue. Biomed Pharmacother 111: 1376-1382, 2019.
21. Zhou YT, Li SS, Ai M, Chen H, Liu YX, Li BY, Zhao Y, Cai WW, Hou B, Ni LL, *et al*: $1,25(OH)_2D_3$ mitigate cancer-related fatigue in tumor-bearing mice: Integrating network pharmacological analysis. Biomed Pharmacother 128: 110256, 2020.
22. Vichaya EG, Ford BG, Quave CB, Rishi MR, Grossberg AJ and Dantzer R: Toll-like receptor 4 mediates the development of fatigue in the murine lewis lung carcinoma model independently of activation of macrophages and microglia. Psychoneuroendocrinology 122: 104874, 2020.
23. Wu Y, Pi D, Chen Y, Zuo Q, Lin L and Ouyang M: Yifei sanjie pills alleviate chemotherapy-related fatigue by reducing skeletal muscle injury and inhibiting tumor growth in lung cancer mice. Evid Based Complement Alternat Med 2022: 2357616, 2022.
24. Kim A, Rivera S, Shprung D, Limbrick D, Gabayan V, Nemeth E and Ganz T: Mouse models of anemia of cancer. PLoS One 9: e93283, 2014.
25. Vichaya EG, Vermeer DW, Budac D, Lee A, Grossberg A, Vermeer PD, Lee JH and Dantzer R: Inhibition of indoleamine 2,3 dioxygenase does not improve cancer-related symptoms in a murine model of human papilloma virus-related head and neck cancer. Int J Tryptophan Res 12: 1178646919872508, 2019.
26. Grossberg AJ, Vichaya EG, Gross PS, Ford BG, Scott KA, Estrada D, Vermeer DW, Vermeer P and Dantzer R: Interleukin 6-independent metabolic reprogramming as a driver of cancer-related fatigue. Brain Behav Immun 88: 230-241, 2020.
27. Grossberg AJ, Vichaya EG, Christian DL, Molkentine JM, Vermeer DW, Gross PS, Vermeer PD, Lee JH and Dantzer R: Tumor-associated fatigue in cancer patients develops independently of IL1 signaling. Cancer Res 78: 695-705, 2018.
28. Zhu X, Burfeind KG, Michaelis KA, Braun TP, Olson B, Pelz KR, Morgan TK and Marks DL: MyD88 signalling is critical in the development of pancreatic cancer cachexia. J Cachexia Sarcopenia Muscle 10: 378-390, 2019.
29. Delitto D, Judge SM, Delitto AE, Nosacka RL, Rocha FG, DiVita BB, Gerber MH, George TJ Jr, Behrns KE, Hughes SJ, *et al*: Human pancreatic cancer xenografts recapitulate key aspects of cancer cachexia. Oncotarget 8: 1177-1189, 2017.
30. Yang S, Yang Y, Chen C, Wang H, Ai Q, Lin M, Zeng Q, Zhang Y, Gao Y, Li X and Chen N: The anti-neuroinflammatory effect of fuzi and ganjiang extraction on LPS-induced BV2 microglia and its intervention function on depression-like behavior of cancer-related fatigue model mice. Front Pharmacol 12: 670586, 2021.
31. Wolff BS, Allen HR, Feng LR and Saligan LN: BDNF Val66Met polymorphism reduces the fatigue-like effects of 5-fluorouracil on voluntary wheel-running activity in mice. Front Behav Neurosci 16: 880969, 2022.
32. Dougherty JP, Wolff BS, Cullen MJ, Saligan LN and Gershengorn MC: Taltirelin alleviates fatigue-like behavior in mouse models of cancer-related fatigue. Pharmacol Res 124: 1-8, 2017.
33. Mahoney SE, Davis JM, Murphy EA, McClellan JL, Gordon B and Pena MM: Effects of 5-fluorouracil chemotherapy on fatigue: Role of MCP-1. Brain Behav Immun 27: 155-161, 2013.
34. Mahoney SE, Davis JM, Murphy EA, McClellan JL and Pena MM: Dietary quercetin reduces chemotherapy-induced fatigue in mice. Integr Cancer Ther 13: 417-424, 2014.
35. Hsu TH, Hung SW, Wu CY, Chiu CC, Hong HT, Lee GC, Chen CC, Lin JS and Wu CP: Supplementation of beef extract improves chemotherapy-induced fatigue and toxic effects in mice. J Funct Foods 75: 104232, 2020.

36. Feather CE, Lees JG, Makker PGS, Goldstein D, Kwok JB, Moalem-Taylor G and Polly P: Oxaliplatin induces muscle loss and muscle-specific molecular changes in mice. Muscle Nerve 57: 650-658, 2018.
37. Sakai H, Sagara A, Arakawa K, Sugiyama R, Hirosaki A, Takase K, Jo A, Sato K, Chiba Y, Yamazaki M, *et al*: Mechanisms of cisplatin-induced muscle atrophy. Toxicol Appl Pharmacol 278: 190-199, 2014.
38. Scott KS, Chelette B, Chidomere C, Phillip West A and Dantzer R: Cisplatin decreases voluntary wheel-running activity but does not impair food-motivated behavior in mice. Brain Behav Immun 111: 169-176, 2023.
39. Chelette B, Chidomere CL and Dantzer R: The GDF15-GFRAL axis mediates chemotherapy-induced fatigue in mice. Brain Behav Immun 108: 45-54, 2023.
40. Gilliam LAA, Moylan JS, Callahan LA, Sumandea MP and Reid MB: Doxorubicin causes diaphragm weakness in murine models of cancer chemotherapy. Muscle Nerve 43: 94-102, 2011.
41. Mackay AD, Marchant ED, Louw M, Thomson DM and Hancock CR: Exercise, but not metformin prevents loss of muscle function due to doxorubicin in mice using an in situ method. Int J Mol Sci 22: 9163, 2021.
42. Zombeck JA, Fey EG, Lyng GD and Sonis ST: A clinically translatable mouse model for chemotherapy-related fatigue. Comp Med 63: 491-497, 2013.
43. Wang Y, van Beurden AW, Tersteeg MMH, Michel S, Kastelein A, Neefjes J, Rohling JHT, Meijer JH and Deboer T: Internal circadian misallignment in a mouse model of chemotherapy induced fatigue. Brain Behav Immun 115: 588-599, 2024.
44. Wang Y, Zanden SYV, van Leerdam S, Tersteeg MMH, Kastelein A, Michel S, Neefjes J, Meijer JH and Deboer T: Induction of fatigue by specific anthracycline cancer drugs through disruption of the circadian pacemaker. Cancers (Basel) 14: 2421, 2022.
45. Bian Y, Sun M, Silver M, Ho KKL, Marchionni MA, Caggiano AO, Stone JR, Amende I, Hampton TG, Morgan JP and Yan X: Neuregulin-1 attenuated doxorubicin-induced decrease in cardiac troponins. Am J Physiol Heart Circ Physiol 297: H1974-H1983, 2009.
46. Fu X, Kong L, Tang M, Zhang J, Zhou X, Li G, Wang H and Fu F: Protective effect of ocotillol against doxorubicin-induced acute and chronic cardiac injury. Mol Med Rep 9: 360-364, 2014.
47. Ray MA, Trammell RA, Verhulst S, Ran S and Toth LA: Development of a mouse model for assessing fatigue during chemotherapy. Comp Med 61: 119-130, 2011.
48. Loman BR, Jordan KR, Haynes B, Bailey MT and Pyter LM: Chemotherapy-induced neuroinflammation is associated with disrupted colonic and bacterial homeostasis in female mice. Sci Rep 9: 16490, 2019.
49. Grant CV, Jordan K, Seng MM and Pyter LM: Antibiotic treatment inhibits paclitaxel chemotherapy-induced activity deficits in female mice. PLoS One 18: e0284365, 2023.
50. Chaillou T, McPeek A and Lanner JT: Docetaxel does not impair skeletal muscle force production in a murine model of cancer chemotherapy. Physiol Rep 5: e13261, 2017.
51. Crouch ML, Knowels G, Stuppard R, Ericson NG, Bielas JH, Marcinek DJ and Syrjala KL: Cyclophosphamide leads to persistent deficits in physical performance and in vivo mitochondria function in a mouse model of chemotherapy late effects. PLoS One 12: e0181086, 2017.
52. Smith LB, Leo MC, Anderson C, Wright TJ, Weymann KB and Wood LJ: The role of IL-1β and TNF-α signaling in the genesis of cancer treatment related symptoms (CTRS): A study using cytokine receptor-deficient mice. Brain Behav Immun 38: 66-76, 2014.
53. Weymann KB, Wood LJ, Zhu X and Marks DL: A role for orexin in cytotoxic chemotherapy-induced fatigue. Brain Behav Immun 37: 84-94, 2014.
54. Wong J, Tran LT, Lynch KA and Wood LJ: Dexamethasone exacerbates cytotoxic chemotherapy induced lethargy and weight loss in female tumor free mice. Cancer Biol Ther 19: 87-96, 2018.
55. Halle JL, Counts BR, Zhang Q and Carson JA: Short duration treadmill exercise improves physical function and skeletal muscle mitochondria protein expression after recovery from FOLFOX chemotherapy in male mice. FASEB J 36: e22437, 2022.
56. Halle JL, Counts BR, Zhang Q, James KM, Puppa MJ, Alway SE and Carson JA: Mouse skeletal muscle adaptations to different durations of treadmill exercise after the cessation of FOLFOX chemotherapy. Front Physiol 14: 1283674, 2023.
57. Barreto R, Waning DL, Gao H, Liu Y, Zimmers TA and Bonetto A: Chemotherapy-related cachexia is associated with mitochondrial depletion and the activation of ERK1/2 and p38 MAPKs. Oncotarget 7: 43442-43460, 2016.
58. Abulizi A, Hu L, Ma A, Shao FY, Zhu HZ, Lin SM, Shao GY, Xu Y, Ran JH, Li J, *et al*: Ganoderic acid alleviates chemotherapy-induced fatigue in mice bearing colon tumor. Acta Pharmacol Sin 42: 1703-1713, 2021.
59. Park HJ, Shim HS, Kim JY, Kim JY, Park SK and Shim I: Ginseng purified dry extract, BST204, improved cancer chemotherapy-related fatigue and toxicity in mice. Evid Based Complement Alternat Med 2015: 197459, 2015.
60. Wang Y, Zhou W, Lyu C, Li Q, Kou F, Jiang M and Wei H: Metabolomics study on the intervention effect of radix salviae miltiorrhizae extract in exercise-induced exhaustion rat using gas chromatography coupled to mass spectrometry. J Chromatogr B Analyt Technol Biomed Life Sci 1178: 122805, 2021.
61. Lu Y, Ning H, Jiang X, Yang R, Song D and Yuan H: Metabolomics reveals hippocampal metabolic fluctuations of postoperative fatigue syndrome and anti-fatigue effect of Carthamus tinctorius L extract in rat model. Biomed Chromatogr 30: 1052-1058, 2017.
62. Yoshizawa K, Kurono R, Sato H, Ishijima E, Nasu H, Ferdaos N, Suzuki H and Negishi K: Effect of sucrose on cisplatin-induced fatigue-like behavior in mice: Comparison with fructose and glucose. Cancer Diagn Progn 1: 95-102, 2021.
63. Ouyang MZ, Lin LZ, Lv WJ, Zuo Q, Lv Z, Guan JS, Wang ST, Sun LL, Chen HR and Xiao ZW: Effects of the polysaccharides extracted from Ganoderma lucidum on chemotherapy-related fatigue in mice. Int J Biol Macromol 91: 905-110, 2016.
64. Wood LJ, Nail LM, Perrin NA, Elsea CR, Fischer A and Druker BJ: The cancer chemotherapy drug etoposide (VP-16) induces proinflammatory cytokine production and sickness behavior-like symptoms in a mouse model of cancer chemotherapy-related symptoms. Biol Res Nurs 8: 157-169, 2006.
65. Lv Z, Liu R, Su K, Gu Y, Fang L, Fan Y, Gao J, Ruan X and Feng X: Acupuncture ameliorates breast cancer-related fatigue by regulating the gut microbiota-gut-brain axis. Front Endocrinol (Lausanne) 13: 921119, 2022.
66. Rafaih AB and Ari K: Artificial intelligence-driven approaches to managing surgeon fatigue and improving performance. Cureus 16: e75717, 2024.
67. Pattanshetty R, Huddar V and Kharwandikar P: Pulmonary function tests, fatigue levels and functional capacity in head and neck cancer patients undergoing cancer therapy: An evaluation study. BLDE Univ J Health Sci 7: 241-244, 2022.
68. Vichaya EG, Darpolor JK, Gross PS, Molkentine JM, Vermeer DW, Vermeer PD, Lee JH, Taniguchi CM and Dantzer R: Associative learning contributes to the persistence of fatigue-like behavior in male mice in a model of cancer survivorship. Brain Behav Immun 107: 296-304, 2023.
69. Vichaya EG, Vermeer DW, Christian DL, Molkentine JM, Mason KA, Lee JH and Dantzer R: Neuroimmune mechanisms of behavioral alterations in a syngeneic murine model of human papilloma virus-related head and neck cancer. Psychoneuroendocrinology 79: 59-66, 2017.
70. Vichaya EG, Ford BG, Moltenkine JM, Taniguchi CM, Phillip West A and Dantzer R: Sex differences in the behavioral and immune responses of mice to tumor growth and cancer therapy. Brain Behav Immun 98: 161-172, 2021.
71. Phan TT, Scott KS, Chelette B, Phillip West A and Dantzer R: The fatigue-inducing effects of cancer and its therapy are characterized by decreased physical activity in the absence of any motivational deficit. Brain Behav Immun 117: 205-214, 2024.
72. Wang Y, Liu Y, Zhang Y, Huo Z, Wang G, He Y, Man S and Gao W: Effects of the polysaccharides extracted from Chinese yam (Dioscorea opposita Thunb.) on cancer-related fatigue in mice. Food Funct 12: 10602-10614, 2021.
73. Zhang X, Lee WD, Leitner BP, Zhu W, Fosam A, Li Z, Gaspar RC, Halberstam AA, Robles B, Rabinowitz JD and Perry RJ: Dichloroacetate as a novel pharmaceutical treatment for cancer-related fatigue in melanoma. Am J Physiol Endocrinol Metab 325: E363-E375, 2023.
74. Chen Z, Wang M, Gao S, Guo H, Wang G and Zhou G: Cardiotoxicity of anti-PD-L1 antibody and the effect of levothyroxine in attenuating the related mortality in mice. Zhongguo Fei Ai Za Zhi 24: 394-403, 2021 (In Chinese).

75. Cao Z, Zhang Y, Jia H, Sun X, Feng Y, Wu H, Xu B and Wei Z: Immune checkpoint inhibitors mediate myocarditis by promoting macrophage polarization via cGAS/STING pathway. Cytokine 187: 156873, 2025.
76. Wolff BS, Raheem SA and Saligan LN: Comparing passive measures of fatigue-like behavior in mice. Sci Rep 8: 14238, 2018.
77. Wolff BS, Renner MA, Springer DA and Saligan LN: A mouse model of fatigue induced by peripheral irradiation. J Vis Exp: 55145, 2017.
78. Renner M, Feng R, Springer D, Chen MK, Ntamack A, Espina A and Saligan LN: A murine model of peripheral irradiation-induced fatigue. Behav Brain Res 307: 218-226, 2016.
79. McDonald TL, Hung AY, Thomas CR and Wood LJ: Localized external beam radiation therapy (EBRT) to the pelvis induces systemic IL-1beta and TNF-alpha production: Role of the TNF-alpha signaling in EBRT-induced fatigue. Radiat Res 185: 4-12, 2016.
80. Wood LJ, McDonald T, Roberts D, Han X, Hung A and Thomas CR: Pelvic irradiation induces a systemic TNF-α response and sickness syndrome in mice: Implications for cancer treatment related fatigue. Int J Radiat Oncol Biol Phys 72 (Suppl): S701-S702, 2008.
81. Huang YM, Chi CW, Wu PS, Tai HC, Chien MN and Chen YJ: Adrenal gland irradiation causes fatigue accompanied by reactive changes in cortisol levels. J Clin Med 11: 1214, 2022.