

HUMAN PHYTOREHABILITATION

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PROTECTIVE ACTION OF ESSENTIAL OILS IN CASE OF ANIMAL IRRADIATION AND POSSIBLE APPLIANCE FOR HUMAN**Tikhomirov Aleksandr Aleksandrovich, Govoroon Mariya Ivanovna**

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Introduction

Protection of human body from ionizing radiation and preventive measures in radiation safety gain a great importance because of anthropogenic disasters and more active appliance of ionizing radiation source in medical practice. That's why it's urgent to search harmless means, which are capable to enhance body resistance to ionizing radiation.

Essential oils (EO) present a wide range of biological activity, which was described in this study: antioxidant properties [14, 15], ability to stimulate immune system [2, 3, 8], healing of wound and burn surface, processes of somatic cell division [1, 10], that is properties, extremely important in case of treatment the radiation injury

Study objective is to investigate if it's possible to decrease injurious action of γ -irradiation due to inhalation with essential oils.

Objects and research methods

Investigations of EO radioprotective properties were carried out according to standard scheme.

Total irradiation of animals was made by X-ray machine RUM-17, KFR 62 sm, combined filter (0,25mm Cu + 1,0mm Al + 0,4mm Sn), dosage rate 0,32-0,42 Gy/min, irradiation dose 7Gy.

The main parameter of radiation injury was mortality percentage of animals for 30 days (%30) after X-ray irradiation. Total characteristic of death dynamics consisted of three stages: death rate of animals from 1-7 days (%7), from 8-14 (%14) and from 15 -30 (%21). On first stage (from the 1st to 7th day) animals die of gastrointestinal tract injury, on the second stage - irreversible damage of blood-making system is a reason of death. Average life of animals (AL), intensity of radiation sickness and rate of death are integral indices of body resistance.

Radiomodifying effect of EO was determined due to value FDV (factor of dosage variation), which reflects decreasing of effective irradiation dosage for animals after preventive procedures and defines their radioprotective action.

Course term (5-10 days or 30 days) and EO dosage were considered while investigating radiopreventive action of EO.

Test animals were placed into closed cells, where content of EO evaporation reached 20 or 100 mg/m³ of air. Practical EO concentrations didn't provoke toxic injury of internals.

Mice breathed air with EO content for 40 minutes every day. Procedure was finished 1 day before irradiation. Control animals were irradiated without EO procedure. In experiments 750 mice (line *CBA/lac*, hybrids (*CBAxC57Bl*) *F2* and outbred white mice weighted 20-25g) were used, 26-30 animals per each group.

Immunogenesis was studied on the model of primary immunal response to T-dependent antigen (erythrocytes of a sheep were injected v/v 5×10^8 cell/ml); type of reaction was assessed by number of antibody-forming cells (AFC) in spleen [13].

Results and discussion

Protective action of inhalation with eucalyptus EO, dose 20 mg/m³.

It was found out that 10 days preventive course of eucalyptus EO caused survival rate of 53,8% against 7,7% of control patterns (average life 47,3 and 14,0 days respectively). FDV was $1,59 \pm 0,33$. Prolongation of EO preventive course up to 30 procedures didn't cause any considerable changes of radio-resistance.

5-days eucalyptus EO course resulted less radioprotective effect: survival rate was 32% of mice while all control specimens died. Average life of "protected" mice made 25,7 days, in control group - 8,5 days.

In this experiment 10-days preventive course of EO gave maximum protective effect.

Marked protective effect of EO became apparent in decreasing of animal death rate, (gastrointestinal type): in 7 days all test animals were alive, while death rate in control groups made 22-44% (depending upon mice lot).

By 14th day survival animals, which were protected by EO, were 1,2, 2,2 and 2,3 times more (EO course terms - 5,10, 30 days) than in control groups. Prolongation of preventive course from 5 up to 30 days was accompanied by decreased death rate (marrowy type) 2,2% per day more [5, 6, 7].

Preventive inhalation with monarda and lavender EO favored survival rate of test mice: lavender EO of 20 mg/m³ content (10-days course) - 34,6% (control group - 7,7%); 30-days lavender EO - 38,4% (3,7% - control groups); monarda EO of 100mg/m³ (30-days course) effected considerably on survival rate - 51,5%.

Method of fractional irradiation permits to reveal influence of radioprotector on postradiation rehabilitation.

Influence of eucalyptus EO for 11 days before irradiation (dosage 0,5 Gy/fraction (total dosage 4 Gy) raised acute irradiation-resistance: in 30 days after acute irradiation 33,3% of animals survived (5% in control group), in 60 days - 25,9% (9,7% in control group).

Mechanisms of EO radioprotection might be the followings:

- injury reduction of blood-making system cells and epithelial cover, that is protective effect of essential oils relatively to quickly divided cells;
- activization of regenerative and reparative processes;
- stimulation of all parts in immunal system: cell-bound, humoral immunities and factors of non-specific resistance;
- activation of antyoxydant body systems;
- reduction of postradiation bacterial complications and etc.

Results of EO influence on animals and human permit to consider them as modulators-adaptogens, which raise body resistance and prepare biological object for further impact of extreme factors, that is irradiation.

That's why we checked EO effect on immune and blood-making systems of healthy animals in order to determine resistance level before irradiation.

Animals inhaled eucalyptus EO for 5,10 or 20 days and last day they were immunized by erythrocytes of a sheep. Animals of control groups were immunized only.

Therefore eucalyptus EO favoured increasing of relative and absolute amount of AFC

in spleen. The most marked effect was after 10 and 20 days course, didn't depend upon EO content. Relative and absolute amount of AFC in spleen increased as follows:

Eucalyptus EO content 20mg/m³:

10-days course – 1,7/2,0 times more (relative/absolute amount of cells).

20-days course – 1,9/2,4 times more.

Eucalyptus EO content 100mg/m³:

10-days course – 1,8-2,1 time more

20-days course – 2,0-2,4 times more.

During 5-days course stimulation was not so considerable: relative amount of AFC increased up to 1,3 times (20 mg/m³) and 2 time with more content of EO.

In this experimental scheme eucalyptus essential oil effected on precursors of lymphocytes, which took part in immune response, and on primary stage of inductive phase of immunogenesis. Stable increasing of lysozyme, titre heterophile antibodies and compliment was marked out.

Influence of essential oils on value of radiation injury of blood-making system and character of its renewal in postradiation period

Opportunity to get over radiation sickness mainly depends upon state of blood-making organs – marrow and spleen [11].

10-days course of inhalations with eucalyptus EO (20 mg/m³) caused increasing of myelocariocytes 1,2 time more (P<0,01). Reduction of spleen weight wasn't accompanied by changes of splenocyte amount.

In this experiment influence of essential oil improved blood-making organs function of healthy animals.

Influence of monarda, eucalyptus and lemon tarragon EO (20 mg/m³) on value of radiation damage of blood-making system and character of its renewal in postradiation period was investigated during the course of 10 sessions.

Irradiation of animals by sublethal and lethal dosages (6Gy) provoked dose-dependent reduction of myelocariocytes number. Inhalations with EO favored either keeping of a large number of viable myelocariocytes or activated their proliferation up to amount which was typical for smaller dosage effect. By 30th day after irradiation amount of myelocariocytes of protected animals (inhaled eucalyptus and monarda EO) corresponded to healthy mice parameter; irradiated animals had the same value 1,6 time less. Preventive action of lemon tarragon was not so expressed: amount of myelocariocytes became 1,4 time more in comparison with parameters of irradiated animals, but didn't reach the level of healthy mice. Effect of EO wasn't registered applying higher doses (6 Gy).

Therefore in a definite dose range EO favors strengthening of reparative processes in blood-forming organs.

Effect of 20-days course of eucalyptus EO exposure (4 Gy) on the cellular composition of marrow and spleen was investigated in dynamics, in 10, 20, 30 and 60 days after irradiation. In 10 days marrow cellularity parameters of "protected" by EO and "unprotected" animals was similar. By 20th day an amount of myelocariocytes was 1,4 time more and by 30th day – 1,5 time more than it was registered in control group. Dynamics of spleen cellularity renewal had the same tendency: by 20-30th days an amount of cells of EO "protected" animals was rather less than "unprotected" had, though by 60th day the difference vanished.

Value of radiation injury of blood-making system wasn't effected by EO in this experimental scheme. Though renewal of marrow cell pool of "protected" animals occurred more actively.

In case of higher irradiation dose EO effect became apparent by 10th day: level of

myelocariocytes of “protected” animals was 2,7 and 3,6 times higher (accordingly to 5 and 6,25 Gy). FDV of marrow cellularity reached 1,225 – quite considerable value for natural radioprotectors.

Immune response correction effecting by eucalyptus and tarragon EO on irradiated mice with sublethal dosage

Under influence of ionized radiation immune system of human and animals is one of critical systems [4].

It was found out that preventive course with eucalyptus EO by 10th day favored increasing of spleen AFC 1,7 time more, in 60 days this value was equal to parameter of healthy mice. Concerning group of radiation control in 60 days an amount of AFC made only 65% of standard.

Dynamics of spleen weight changes, its cellularity and an amount of AFC after preventive course with tarragon EO corresponded to the radiation control.

In the event inhalations with eucalyptus and tarragon were carried out after irradiation, in 30 days reduction of absolute number of AFC and a number of splenocytes occurred that renewed up to radiation control by 60th day.

Prevention of endogenous infection applying essential oils

Postradiation period of animals and human is characterized by clinical syndrome with infectious complications.

Antibacterial activity of EO *in vitro* is described in a lot of works of native and foreign authors. *In vitro* essential oils effect in complicated combination with protective body structures, strengthening each other mutually. As a result of this interaction even micro dose of essential oils displays antibacterial and anti-inflammatory action.

10-days course of inhalations with eucalyptus EO (20 mg/m³) was used to prevent endogenous infections in irradiated body. In a day after that course was finished rats *Vistar* were exposed to radiation of lethal dose 8,5 Gy, that can cause animal death of bulks for the first 7 days.

The most of unprotected rats died because of intestinal disorders: in 7 days only 12,5% of rats survived. Inoculations out of histic homogenate of liver and spleen resulted development of microorganisms (100% of animals), multiple growth was registered in a half of cases, that is animals had heavy sepsis.

After eucalyptus EO effect 60% of irradiated animals survived by 7th day; rate and intensity of bacterial contamination of internals reduced considerably – 33% of positive cases with no multiple growth. Bacterial contamination of internals of rats, irradiated by dose 8,5 Gy and animals protected by eucalyptus EO was decreasing till the level of animals that were exposed to radiation of 6 Gy.

Otherwise applying of EO eased endogenous infections of animals equally as if irradiation dose became 2,5 Gy less [8].

In conclusion of this serious of experiments it's worth to note that it was found out a class of compounds which possesses radioprotective properties, though it hasn't been applied with this purpose before.

By present there is information that EO can be considered as universal preventive remedy, which possesses a wide range of therapeutic effect: EO is already used to prevent SARS (Acute respiratory syndrome) [9], correction of secondary immunodeficiency [2, 3], also to treat chronic diseases [11, 12, 14, 15], to relief xenobiotic action on human body [10] and etc.

In Yalta KRU “NII named after I.M. Sechenov” essential oils were successfully used for rehabilitation treatment of emergency wreckers at Chernobyl power station with long-term

effect of radiation: as a result considerable improvement of characteristics during laboratory investigations and clinical state of patients were fixed. Schemes of cure included as separate essential oils as mixtures of 3-5 essential oils, duration and concentrations of preparations were worked out.

Children from zone of radionuclide pollutions had pathologic disorders characterized by systemic nature. It became apparent by early progressing disorders of main functional systems: as a rule injuries were poliorganic, chronic nidus of infection and disorders of immune and neuroendocrinal systems were registered as well. Applying of essential oils favored renewal of cell and humoral immunities, minimized imbalance of cytokine cascade and hemoblastoses, reduced rate of attaching respiratory virus infection [5].

Development of nuclear power engineering exposes more and more people to radiation: workers who deals with radiation sources, people exposed to radiation during anthropogenic disasters and sick people who got radio- and X-ray therapy. In these categories of people annual increasing of sickness rate by all main disease classes is actual for people from categories mentioned above.

This data permits to apply EO in treatment of people exposed to radiation.

Conclusions

In terms of this investigation radioprotective properties of compounds, which haven't been used with this purpose before, were revealed. The animal experiment displayed a considerable effect of essential oils relative to blood-making and immune systems, possibility to reduce intensity of septic processes and as a result to prolong animal life after irradiation by lethal dose of γ -ray.

Working as adaptogens, EO favor improvement of critical systems when organism of animals meet radiation being in a state of increased functional activity and protection of blood-making and immune organs.

Including of EO procedures into scheme of rehabilitation treatment of emergency wreckers at Chernobyl power station with long-term effect of radiation improved characteristics of laboratory investigations and clinical state of patients.

This data is a ground for further development in applying of essential oils as preventive meanings for workers who deal with radiation sources and as a remedy for people who was exposed to radiation by case or with therapeutic purpose.

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Animal experiment showed radioprotective effect of essential oils on hematopoietic, immune systems and possible decreasing of septic process rate. Appliance of essential oils in rehabilitation treatment of emergence wreckers at NPS in Chernobyl favored considerable improvement of clinical state of patients and parameters of laboratory investigations.

A class of compounds, which haven't been used specially with that purpose before, possesses radioprotective properties.

Key words: *essential oils, dosages, γ -irradiation, anti-radiation action*