British Journal of Medicine & Medical Research
21(3): 1-7, 2017; Article no.BJMMR.32673
ISSN: 2231-0614, NLM ID: 101570965

SCIENCEDOMAIN *international*
www.sciencedomain.org

New Therapy Strategy for Prostate Cancer: *Amanita phalloides* Treatment Stabilizes Best Without Pre-treatments (Observational Study Pre-protocol)

Isolde Riede[1*]

[1]Independent Cancer Research, Im Amann 7, D-88662 Ueberlingen, Germany.

Author's contribution

The sole author designed, analyzed and interpreted and prepared the manuscript.

Article Information

DOI: 10.9734/BJMMR/2017/32673
Editor(s):
(1) Mahin Khatami, National Cancer Institute (ret.), The National Institutes of Health, Bethesda, MD, USA.
(2) Evangelos Marinos, University of Athens, School of Medicine, Laboratory of Biology, Athens, Greece.
(3) Edward J. Pavlik, University of Kentucky Medical Center, Division of Gynecological Oncology, USA.
Reviewers:
(1) Katia Candido Carvalho, FMUSP Faculdade de Medicina da Universidade de São Paulo, Brazil.
(2) S. Sundaresan, SRM University, Kattankulathur, India.
Complete Peer review History: http://www.sciencedomain.org/review-history/18972

Study Pre-Protocol

Received 9th March 2017
Accepted 4th May 2017
Published 8th May 2017

ABSTRACT

Background: *Amanita phalloides* (Amanita) contains amanitin, inhibiting RNA polymerase II. Partial inhibition with amanitin influences tumor cell - but not normal cell - activity. Patients treated with Amanita often gain a stable disease state without further tumor growth.
Aim: Several therapies for prostate cancer are in use so far. This study evaluates the implementation of the Amanita therapy into the therapeutic regimen of today.
Methods: 38 Patients with diagnosed prostate cancer were treated with Amanita alone. Prostate specific antigen (PSA) was used as a parameter for tumor cell growth. Data about previous therapies like anti androgen treatment, prostatectomy, chemotherapy or radiation and progression state of the disease were collected, and relationships analyzed.
Results: Some patients could be stabilized with Amanita, evidenced by previously increasing PSA that did not increase for at least six months during Amanita therapy. In some patients PSA levels further increased despite Amanita therapy indicating immediate therapy resistance. Analyses revealed that successful Amanita therapy is strongly associated with beginning therapy early, while progressive disease states are often resistant to Amanita (*p*-value 0,005). Correlation between

**Corresponding author: E-mail: riede@tumor-therapie.info;*

tumor specific pre-treatments and Amanita therapy resistance is significant: all patients without pre-treatment could be stabilized with Amanita (*p*-value 0,007).
Conclusions: Amanita therapy can reduce tumor growth best in patients without previous treatments, and was most effective in patients without tumor progression. Therefore Amanita should be used first as a tumor specific therapy. Anti androgen treatment, chemotherapy, radiation or prostatectomy can be applied at later stages.

Keywords: Amanita phalloides; prostate cancer; new tumor therapy strategy.

1. INTRODUCTION

A tumor can develop in any tissue, including the prostate. Different kinds of growth activity and metastasizing activity occur, depending on the origin of proliferation. A slow growing benign prostate cancer can persist without symptoms, while an aggressive malignant tumor may cause difficulty in urinating, problems with sexuality, including erectile dysfunction and pain. Prostate cancer tends to develop in men over the age of fifty. It is the most prevalent type of cancer in men. Diagnosis may be indicated by symptoms, physical examination, PSA screening, or biopsy.

Currently therapy starts at a PSA level of 10 ng/ml. Prostate cancer is influenced by androgens. Most often anti androgen treatments are applied first with variant drugs, or combinations of drugs. 5 alpha-reductase inhibitors are used with variable success since the 1990s [1,2,3]. A maximal androgen blockade is frequently used today [4]. This treatment is accompanied by severe side effects. Despite the maximal blockade resistance will eventually occur, requiring new treatment options [5,6]. After the appearance of resistant cells, identified by increasing PSA levels, chemotherapy or radiation is applied. Diverse immune therapies are in evaluation also.

In a genetic and molecular study with tumor forming Drosophila melanogaster, four classes of genes involved in tumor formation could be distinguished. Proliferative genes disrupt cell cycle control, and one single proliferative mutation is sufficient for tumor formation [7, Review]. Mutations in oncogenes and tumor-suppressor genes are secondary events and exert their effects by destabilization of the differentiation pattern of the cells. In tumor cells, usually a proliferative gene mutation allows replication immediately after mitosis. This forces the cell to divide: An onset of replication leads the cell through a short gap phase and into mitosis again. No molecular point of attack against tumor growth has been identified in this class of genes. The central potential targets for therapeutic intervention are switch genes. These genes can switch the tumor growth ON or OFF. In tumor cells the switch genes are over-expressed [8]. This switches the growth of tumors to ON. All switch genes belong to the class of HOX genes, and use an enzyme for their action: RNA PolymeraseII (RNAP). If the switch genes are over expressed, RNAP is used to full extent, creating a bottleneck for tumor growth. The extract of Amanita contains (alpha)-amanitin, among other active peptides, that lead amanitin into the cellular nucleus, where RNAP works [9, 10]. The phallotoxin volva is a membrane protein, leading amanitin into the cell. Other phallotoxins, amatoxins or virotoxins interact with cellular functions, stimulate polymerizations or stabilize at the level of actin. One molecule of amanitin binds to one molecule of RNAP and destroys its function completely. RNAP in somatic cells has lower levels of activity. Inhibition of 50% of RNAP has no effect on normal cells, but inhibits tumor growth of cells. These tumor cells can be recognized by the immune system and digested. Through this approach it is possible to stabilize the disease state for years.

The treatment with Amanita is monitored with a tumor marker. Amanita inhibits tumor growth of cells, therefore the level of the tumor marker should not further increase. Prostate cancer growth can be monitored with PSA. Increasing PSA reflects an increasing number of cells. Patients with prostate cancer have been treated successfully for years with Amanita, which stabilized PSA levels, halting both, progression and symptoms [11]. After years of therapy, higher doses of the drug were necessary for stabilization. To overcome resistance, intervals with other therapies were able to re-establish sensitivity to the drug [12]. Here a clinical study with prostate cancer patients is presented. At various stages, patients were treated exclusively with Amanita. The results were summarized and statistically evaluated.

2. METHODS

Amanita phalloides is used since 300 years, the classical indication is fear of death. In molecular sense Amanita was newly interpreted to be able to inhibit specifically the growth of tumor cells. After a complete anamnesis the patients were treated with *Amanita phalloides* (zert. Riede) D2 [Herbamed AG, CH], at an average dose of 4 x 10 drops per day, resulting in an average uptake of 50 ml per month. The daily dose contains 50 molecules of amanitin per cell. With 100 ml of this drug, about 50% of all RNAP molecules in all cells are inhibited. Degradation of the drug is unknown. Amanita breaks the activity of the tumor cells without affecting somatic cells. Usually no side effects occur in patients with small tumors. In patients with large tumors, the therapy can cause pain, due to the degradation of tumor cells. In those cases the dose is reduced to an amount where the pain is minimized by a maximal possible degradation. Degradation of cells is monitored with lactate dehydrogenase levels in serum. Monitoring of PSA and lactate dehydrogenase regularly shows the optimal minimal dose for stabilization of the disease.

Amanita can be used to inhibit growth of all type of tumors. Successful treatments of Mammary-carcinoma, thyroid cancer, colon cancer or B cell carcinoma have been described [7]. Monitoring is necessary to optimize the dosage and to avoid resistance development. Amanita should not be applied without a diagnosed tumor, otherwise a later appearing tumor could develop resistance against the drug. In a molecular sense Amanita has been found to be able to specifically inhibit the growth of tumor cells [7]. In all patients, in addition to Amanita as the only tumor specific drug, the oral uptake of essential fatty acids and dermal application of Zinc salve was indicated. Essential fatty acids enhance the fluidity of cellular membranes, and decrease the risk of autoimmunity. For monitoring of the therapy, the regular measurement of PSA was performed. The minimal follow up was for 6 months. Therapy details have been reported [11,12].

3. RESULTS

Out of 69 patients with prostate cancer, 31 were excluded from this study because of missing information and non-compliance. Most frequently they used the Amanita therapy only for a few weeks. 38 patients were selected that used the Amanita therapy for at least six months.

The patients were classified into two categories, first those with a positive response to the Amanita therapy consisted of a previously increasing PSA that was stabilized due to the drug. Each patient was categorized with regard to age, metastases, pre-treatments and familial risk as outlined in Table 1.

The second class of patients was composed of weak therapy responders consisting of an increasing PSA that was not stabilized by Amanita. These patients were ordered into the same categories as outlined in Table 1.

Data was limited in many categories and did not allow statistical evaluation. Therefore in most cases only tendencies are described. Three categories characterized a high imbalance between the Amanita responders and Amanita non responders: Responders consisted of:

- 100% of patients without any tumor specific pre-treatment
- 84 % of patients without metastases
- 71 % of patients without anti androgen pre-treatment.

To evaluate the significance of these relationships, the Pearson Chi-Square was utilized (Table 2). The correlation between absence of metastases and response to Amanita resulted in a high Pearson score (10,795, *p*-value 0,005). Response to Amanita was also correlated in patients without any tumor specific pre-treatment (Pearson score = 9,871, *p*-value 0,007). Resistance to Amanita in patients with anti androgen pre-treatment was high but not significantly correlated with response to Amanita treatment (Pearson score = 3,527, *p*-value 0,171). Response to Amanita also correlated elder age and lower Gleason score. There was no correlation with familial risk.

4. DISCUSSION

Significantly all patients without tumor specific pre-treatment could be stabilized with Amanita. Consequently pre-treatment can cause immediate resistance to Amanita. How can this be understood?

The natural standard mutation rate occurs at a rate of $1{:}10^6$. Without selective pressure, most of these mutations remain without consequence, no advantage or disadvantage for the cell exists. Due to a selective pressure some of these mutations can cause resistance, the cell has an

advantage with this mutation and overgrows the cells without mutation. With this overgrowth, resistance to a selective pressure from anti androgens can occur within weeks. This is usually the case when using a single anti androgenic drug (example in [12]).

A maximal androgen blockade using non steroidal anti androgen plus medical or surgical castration, blocks hormone production at different biochemical targets in androgen metabolism. This induces a high selective pressure for cellular resistance, which occurs after several years [13].

Figs. 1 to 4 illustrate several situations in different therapy strategies. A mutation, that naturally occurs, will not have an advantage, if no selective pressure is present. Therefore, the cell with the mutation has no advantage, and will not

Table 1. Data

	No. of Patients 1)	Responders 2)	Non Responders 3)	Mean 4)
Data used from	38	22	16	58%
Age below 50 5)	3	2	1	67%
Age 50-59	8	4	4	50%
Age 60-69	17	9	8	53%
Age above 69	11	8	3	73%
No metastasis	19	16	3	**84%**
Metastases	19	6	13	32%
Gleason 6-7	8	6	2	75%
Gleason 8-9	6	3	3	50%
No pre-treatment	10	10	0	**100%**
Pre-treatments	28	12	16	43%
After radiation	6	1	5	17%
After chemotherapy	5	1	4	20%
After prostatectomy	13	7	6	54%
After anti androgen	17	7	10	41%
No anti androgen	21	15	6	**71%**
Familial risk	17	8	9	47%
No familial risk	4	3	1	75%

1) 31 patients were excluded because of lack of information. Given are the number of patients fulfilling the criteria 2) Patients that showed an increasing PSA before the Amanita therapy and a stable PSA during at least 6 months under Amanita therapy. 3) Patients that showed an increasing PSA before the Amanita therapy and an increasing PSA during Amanita therapy. 4) Given is % of stable patients fulfilling the criteria. 5) Age of patient at beginning of Amanita therapy

Table 2. Relationship

	Responders	Non Responders	
Metastases			SUM
No	16	3	19
Yes	6	13	19
SUM	22	16	38
Pearson Chi-Square 1)		**10.795**	very significant
All tumor specific therapies			
No	10	0	10
Yes	12	16	28
SUM	22	16	38
Pearson Chi-Square		**9.871**	very significant
Anti androgen therapies			
No	15	6	21
Yes	7	10	17
SUM	22	16	38
Pearson Chi-Square		**3.527**	not significant

1. Pearson Chi-Square in excess of the critical value 3,841 show significant relationship between parameters. Explanations in the Text

overgrow the cells without mutation (Fig. 1). If a weak selective pressure is present, a natural mutation can lead to resistance. In this case, the mutant cell has a small advantage over non mutant cells, and will slowly overgrow them. If the mutant cell finds a way to overcome a high selective pressure, the advantage over the cells without mutation is high and overgrowth is fast.

Fig. 2 indicates a therapy avoiding resistance. A therapy inducing weak selective pressure leads to resistance. In a period with no selective pressure, the mutant cells have no further advantage over the non mutant cells. This might lead to overgrowth of the cells without mutation. Furthermore, a high selective pressure induces faster resistance development. In the following interval without selective pressure the cells with the mutation causing resistance cannot vanish completely.

Fig. 3 shows the situation of an ideal therapy. Two drugs, acting at different biochemical targets, each sufficient to hinder disease progression are used in alternating intervals. With drug 1 a light selective pressure leads to a slow resistance appearance. After this is measurable, the drug 1 is replaced by drug 2. By applying a low selective pressure by drug 2, the resistance to drug 1 can vanish. Alternating intervals can keep the disease stable for a long time.

Fig. 4 shows how to induce resistances. With several drugs given simultaneously, resistances appear that are difficult to overcome with alternative therapy strategies. This is seen in Amanita non responders: pre-treatments already lead to resistance against other drugs. All tumor therapies that impose strong selective pressure will end in resistance development. All cellular functions interfere with biochemical pathways. Each resistance is due to a mutation in a gene of a cell. Multiple resistances are consequently due to multiple mutations in different genes. The more mutations occur, the more biochemical pathways are affected. In the end, all resistances interfere with normal cell function.

Years after therapy with *Amanita phalloides* the drug still is able to restain tumor cell growth. This might be due to the low concentration of the drug: it is applied not to poison the tumor cell, but to reprogram the molecular switch leading to tumor formation. Therefore only a weak selective pressure is induced here: the average dose per day inhibits 50 of the 15,000 RNAP molecules per cell by binding it irreversibly. Anti androgens are applied at higher doses: 5-alpha reductase inhibitors are applied in a dose of 15,000 molecules per cell, and the androgen receptor is inhibited with 1,000,000 flutamide molecules per cell. Higher exposures per cell increase the selective pressure.

Amanita therapy shows no side effects. The quality of life is unaffected. All other tumor therapies induce severe side effects.

A strategy in tumor therapy should consider that all tumor cells are human somatic cells. They all have the same human cell structure. There are no cell structures in tumor cells – like in bacteria – that are not present in other somatic cells. A tumor therapy should not use drugs or physical methods that also destroy normal body cells. A tumor therapy should therefore use weak selective pressure to avoid resistance development. Only one drug at a time, in a minimal dose, for a maximal duration, should be

Fig. 1. If no selective pressure is present, cells with mutations have no advantage and cannot overgrow non mutant cells (red arrow). With a weak selective pressure (blue arrows), mutant cells with resistance against it have a small advantage over non mutant cells and overgrow them slowly. Under a strong selective pressure, the advantage of mutant resistant cells is high, and they overgrow the non mutant cells fast

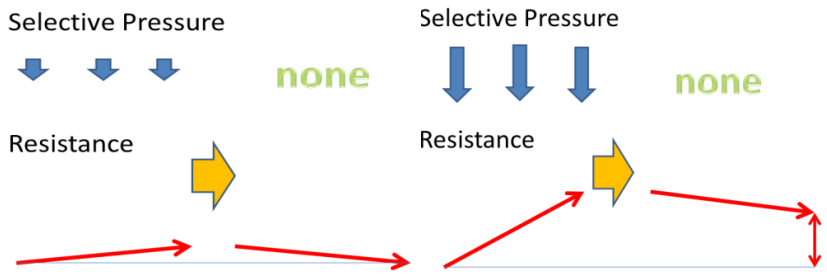

Fig. 2. Weak selective pressure (blue arrows) leads to slow overgrowth of mutant resistant cells (red arrow). In a following interval with no selective pressure (yellow arrow) no advantage of the resistant cells occurs, they vanish slowly. Strong selective pressure leads to a fast resistance appearance. In the following period with no selective pressure resistant cells will not vanish completely

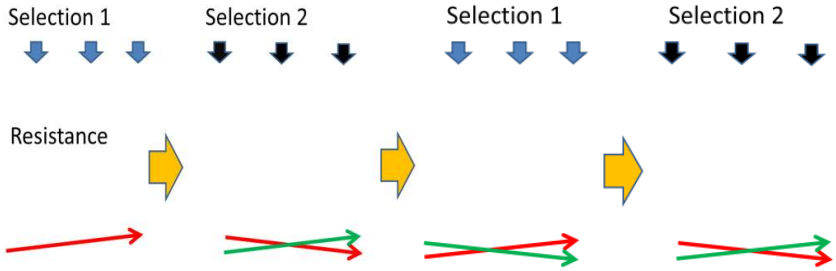

Fig. 3. Long term stabilization by alternating interval therapies: Weak selective pressure 1 (blue arrows) is followed (yellow arrow) by weak selective pressure 2 (black arrows). During this phase the resistance to selective pressure 1 can vanish (red arrows), and resistance against selective pressure 2 appears (green arrows)

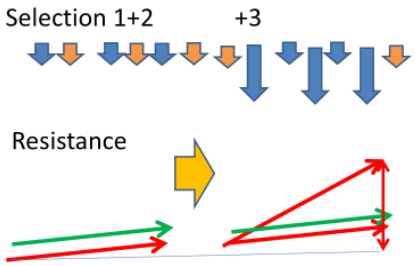

Fig. 4. Inducing resistance: By applying several selective pressures simultaneously inevitably resistance develops that cannot be controlled any more

used. This tactic requires monitoring of the disease state. In case of resistance, an interval with another drug treatment that focuses on a different biochemical target, again with a low selective pressure should be used. In this interval the resistance against the first drug can disappear.

5. CONCLUSION

Amanita reprograms the tumor cell and reduces tumor cell activity. This therapy can be given in advance of other tumor therapies. The patient should be able to receive treatments without severe side effects.

CONSENT

As per international standard or university standard, patient's written consent has been collected and preserved by the author's.

ETHICAL APPROVAL

Only existing drugs in usual doses are used during this study. No new drug was applied.

Therefore no ethical permission is necessary in this case.

COMPETING INTERESTS

Author has declared that no competing interests exist.

REFERENCES

1. Gormley GJ. Role of 5 alpha-reductase inhibitors in the treatment of advanced prostatic carcinoma. Urol Clin North Am. 1991;18(1):93-8.
2. Yin T, Qiao Z, Li Y, Li D, Jiang M, An C, Wang F, Zuo M, Hu K, Li Q. Comparisons of the efficacy and safety of finasteride and dutasteride for benign prostatic hyperplasia: A network meta-analysis. Am J Ther.; 2015.
3. Tammela TL. Endocrine prevention and treatment of prostate cancer. Mol Cell Endocrinol. 2012;360(1-2):59-67.
DOI: 10.1016/j.mce.2012.03.002
Epub 2012
4. Angulo JC, Redondo C, Sánchez-Chapado M, Colás B, Ropero S, López JI. Survival predictors in patients with prostate adenocarcinoma with hormonal blockade. Pathol Res Pract. 2016;2(10):899-903.
DOI: 10.1016/j.prp.2016.07.012
5. Flüchter SH, Weiser R, Gamper C. The role of hormonal treatment in prostate cancer. Recent Results Cancer Res. Urology. 2007;175:211-37.
(2017 Mar 13)
pii: S0090-4295(16)31023-8.
DOI: 10.1016/j.urology.2016.12.033
6. Crawford ED, Petrylak DP, Shore N, Saad F, Slovin SF, Vogelzang NJ, Keane TE, Koo PJ, Gomella LG, O'Sullivan JM, Tombal B, Concepcion RS, Sieber P, Stone NN, Finkelstein SE, Yu EY. The role of therapeutic layering in optimizing treatment for patients with castration-resistant prostate cancer (RADAR II). Urology; 2017.
(2017 Mar 13).
pii: S0090-4295(16)31023-8.
DOI: 10.1016/j.urology.2016.12.033
7. Riede I. Switch the tumor off: From genes to amanita therapy. American Journal of Biomedical Research. 2013;1(4):93-107.
DOI: 10.12691/ajbr-1-4-5
Available:http://www.sciepub.com/ajbr/content/1/4
8. Rozovskaia T, Feinstein E, Mor O, Foa R, Blechman J, Nakamura T, Croce CM, Cimino G, Canaani E. Upregulation of Meis1 and HoxA9 in acute lymphocytic leukemias with the t(4:11) abnormality. Oncogene. 2001;20(7):874-8.
9. Fahrenholz F, Faulstich H, Wieland T. Components of the green deathcap toadstool, *Amanita phalloides*. XLII. Peptide syntheses. 48. Synthesis of norphalloin and of a monocyclic compound with 18-membered ring. [Article in German] Justus Liebigs Ann Chem. 1971;743:83-94.
10. Vetter J. Toxins of *Amanita phalloides*. Toxicon. 1998;36(1):13-24.
11. Riede I. Tumor therapy with *Amanita phalloides* (Death Cap): Long term stabilization of prostate-cancers. J Integr Oncol. 2012;1:101.
DOI: 10.4172/jio.1000101
12. Riede I. Stabilization of prostate cancer with *Amanita phalloides*: Intervals with 5-Alpha-Reductase Inhibitors and Melatonin to Circumvent Resistance. Case Report British Journal of Medicine and Medical Research. 2016;17(5):1-6
DOI: 10.9734/BJMMR/2016/27895
13. Reyes EE, VanderWeele DJ, Isikbay M, Duggan R, Campanile A, Stadler WM, Vander Griend DJ, Szmulewitz RZ. Quantitative characterization of androgen receptor protein expression and cellular localization in circulating tumor cells from patients with metastatic castration-resistant prostate cancer. J Transl Med. 2014;12:313.
DOI: 10.1186/s12967-014-0313-z

Peer-review history:
The peer review history for this paper can be accessed here:
http://sciencedomain.org/review-history/18972