Eur J Nutr 41 : 264 – 270 (2002)
DOI 10.1007/s00394-002-0384-0

ORIGINAL CONTRIBUTION

Bernhard Watzl
Achim Bub
Karlis Briviba
Gerhard Rechkemmer

Acute intake of moderate amounts of red wine or alcohol has no effect on the immune system of healthy men

Summary *Background* A recent prospective cohort study revealed that moderate wine consumption but not consumption of other alcoholic beverages is associated with a decreased risk of common cold. In contrast, wine constituents such as ethanol and polyphenols are known to suppress immunity. *Aim of the study* We investigated whether acute intake of a moderate amount of alcohol modulates immune functions in healthy men and whether polyphenols in red wine with antioxidative and immunomodulatory potential induce changes in immune functions that differ from those induced by the consumption of the 12 % ethanol. *Methods* Six healthy males with moderate alcohol consumption patterns randomly consumed a single dose of 500 ml of red wine (12 % ethanol), a 12 % ethanol dilution, dealcoholized red wine, and red grape juice, respectively. The following immune functions were measured before beverage consumption and 1, 3, and 24 h later: phagocytic activity and intensity of neutrophils and monocytes, production of tumor necrosis factor-alpha, interleukin-2, and interleukin-4, lymphocyte proliferation, and lytic activity of natural killer cells. *Results* Acute consumption of a moderate amount of red wine and of a 12 % ethanol solution had no effect on immune functions in men. Acute consumption of polyphenol-rich beverages (dealcoholized red wine and red grape juice) also did not affect immunity. *Conclusions* This study clearly shows that moderate consumption of alcohol at doses which inversely correlate with cardiovascular disease risk has no short-term effect on human immune cell functions. Acute intake of polyphenol-rich beverages such as red grape juice and dealcoholized red wine also does not affect immunity.

Key words red wine – red grape juice – ethanol – anthocyanins – immune system

Received: 27 August 2002
Accepted: 24 September 2002

Bernhard Watzl, PhD (✉) · A. Bub · K. Briviba · G. Rechkemmer
Institute for Nutritional Physiology
Federal Research Centre for Nutrition
Haid-und-Neu-Str. 9
76131 Karlsruhe, Germany
Tel.: +49-721/6625-410
Fax: +49-721/6625-404
E-Mail:
bernhard.watzl@bfe.uni-karlsruhe.de

Introduction

In epidemiological studies moderate alcohol consumption has been shown to protect people from coronary heart disease (Renaud et al. 1998, Rimm et al. 1999, Gaziano et al. 1999, Gronbæk et al. 2000). Furthermore, moderate intake of alcohol (up to three drinks) in nonsmokers intentionally exposed to rhinoviruses was associated with a decreased risk of the common cold (Cohen et al. 1993) suggesting that alcohol may stimulate the immune response against this type of virus. In a recent prospective cohort study, total alcohol intake, beer and spirits consumption were not related to the occurrence of common cold, whereas moderate consumption of wine was inversely associated with the risk of common cold. The association was stronger for red wine when compared with white wine (Takkouche et al. 2002) indicating that specific polyphenols in red wine contribute to the observed reduction in common cold risk.

In contrast, consuming more than three drinks of alcohol per day is associated with an increased all-cause mortality (Bofetta and Garfinkel 1990). Chronic ethanol abuse is known to result in specific defects in innate and acquired immunity and may increase host susceptibility to infections (Watzl and Watson 1992, Nelson and

EJN 384

Kolls 2002). Recent experimental evidence suggests that acute, moderate alcohol consumption can also affect the immune system. Pharmacological doses of ethanol *in vitro* significantly suppress the production of pro-inflammatory cytokines such as tumor necrosis factor-α (TNFα) in human peripheral blood mononuclear cells (PBMC) (Watzl and Watson 1993, Szabo et al. 1996). Moreover, ethanol exposure of polymorphonuclear neutrophilic granulocytes (PNG) and monocytes *in vitro* and *in vivo* inhibits phagocytosis (Stoltz et al. 1999) and decreases their capacity to produce reactive oxygen species (Patel et al. 1996, Szabo 1999). However, relatively little is known of the effects of acute alcohol consumption on the immune system of humans (Szabo 1999).

Alcohol consumption and its subsequent metabolism in the liver generates reactive oxygen species, which interfere with various immune cell functions. Phenolic antioxidants in red wine may scavenge reactive oxygen species and thereby protect the host against impairment of immune cell functions due to ethanol intake. Based on the French wine consumption of 180 ml/d, the daily intake of polyphenols from wine can be estimated to be as high as 500 mg/person (Teissedre and Landrault 2000). Recently, in an animal study it was shown that ethanol intake in the form of red wine produced no changes in blood lymphocyte and NK cell numbers, while a similar intake of ethanol alone reduced the percentage of these cells (Percival and Sims 2000). This suggests that polyphenols in red wine may prevent toxic effects of high ethanol doses. Immune cell functions, however, were not measured in this study. Red wine contains a variety of polyphenols with strong *in vitro* as well as *in vivo* antioxidant (Wang et al. 1997, Casalini et al. 1999, Lodovici et al. 2001) and immunomodulatory effects (Middleton et al. 2000). Despite this, the immunological effects of flavonoids in humans have not been studied yet. Most polyphenol studies were performed in *in vitro* systems demonstrating that flavonoids suppress a variety of immune functions including lymphocyte proliferation, lytic activity of NK cells, and cytokine secretion (Middleton et al. 2000).

The objective of this study was first to investigate 1) whether the acute consumption of a moderate amount of ethanol modulates immune functions in healthy men during a period of 24 h, and 2) whether polyphenols in red wine with their antioxidative and immunomodulatory potential induce changes in immune functions that differ from those induced by the consumption of a 12 % ethanol beverage.

Subjects and methods

Subjects and study design

Six non-smoking men with moderate alcohol consumption patterns and with normal body weight were recruited for the study. All subjects were in good medical health as determined by a screening history and medical examination. None were taking vitamin supplements or medications two months before or during the study. The study was approved by the Medical Ethical Committee of the Landesärztekammer Baden-Württemberg and all participants gave their consent in writing.

The study was performed under strictly-controlled conditions at the human nutrition unit of the Federal Research Centre for Nutrition within a period of 4 weeks. During the study period, subjects adhered to their usual diets, but were instructed to avoid food products rich in anthocyanins or polyphenols (Bub et al. 2001). Each volunteer had 4 experimental treatments: after an overnight fast, subjects consumed 500 mL of red wine (RW; 12 % ethanol v/v), dealcoholized red wine (DRW), red grape juice (RGJ), or ethanol (ETOH; 12 % ethanol v/v), with a wash-out period of one week between each experimental day. The subjects were randomly assigned to one of the 4 groups; each subject received the beverages in a different order. With the beverages subjects ate 2 white rolls (150 g). Two hundred min after breakfast, subjects consumed a standardized meal, and further meals until the end of the 24 h-study period were provided to all study subjects.

RW (variety Lemberger) and RGJ (variety Lemberger) grown in the same vineyard were obtained from the State Winery Weinsberg (Baden-Württemberg, Germany). Dealcoholization of the RW was achieved by a vacuum rectification process (Center for dealcoholization EAZ Petershans GmbH, Waiblingen, Germany). The anthocyanin (mg/L) and catechin (mg/L) contents were 171.1 and 63.4 (RW), 144.8 and 47.0 (DRW), and 338.6 and 64.3 (RGJ), respectively. The 12 % ethanol beverage was prepared from vodka.

Collection and preparation of blood samples

Fasting venous blood samples were collected before, 1, 3, and 24 h after beverage consumption. Blood was drawn from an antecubital vein into prechilled tubes containing Li-heparin (Monovette-Sarstedt, Nümbrecht, Germany) and immediately placed on ice in the dark. Plasma was collected after centrifugation at 1500 x g for 10 min at 4 °C. Tubes without anticoagulant (Serum-Monovette-Sarstedt, Nümbrecht, Germany) were used for serum collection. Blood was allowed to clot at RT for 30 min, then centrifuged at 1500 x g for 10 min at RT. The serum was stored at –80 °C until analysis.

Peripheral blood mononuclear cells (PBMC) were isolated by density gradient centrifugation using Histopaque 1077 (Sigma, Deisenhofen, Germany) and resuspended in complete RPMI-1640 culture medium (Life Sciences, Eggenstein-Leopoldshafen, Germany), containing 5 % (v/v) heat-inactivated FBS (Life Sciences), L-glutamine (2 mmol/L), penicillin (100,000 U/L) and streptomycin (100 mg/L).

Blood ethanol concentration. Blood ethanol concentrations were measured using a commercial kit (Hoffmann-LaRoche, Mannheim, Germany).

Lymphocyte proliferation. PBMC at 1 x 10^9 cells/L in medium containing 10 % of FBS were stimulated by the T cell mitogen concanavalin A (5 mg/L, ConA, Sigma, Deisenhofen, Germany) for 120 h at 37 °C. Proliferation was measured using the pyrimidine analogue 5-bromodeoxyuridine, which was quantified in PBMC by a cellular enzyme immunoassay as described earlier (Watzl et al. 2000).

Quantification of cytokine secretion. PBMC at 1 x 10^9 cells/L were cultured in medium containing 10 % of FBS and stimulated by 5 mg/L ConA for 24 h at 37 °C (IL-2, IL-4) or by 1 µg/L LPS (TNFα). Cell-free supernatants were collected and stored at –80 °C until analysis. TNFα, IL-2 and IL-4 were measured by sandwich-ELISAs as described previously (Watzl et al. 2000).

Lytic activity of NK cells. Lytic activity of NK cells against K562 target cells (effector:target ratios 50:1, 25:1, 12.5:1) was measured with the flow cytometric method (Watzl et al. 2000).

Phagocytic activity and intensity. Assessment of phagocytic activity (percentage of phagocytic-active cells) and phagocytic intensity (number of phagocytized *E. coli* expressed as mean fluorescence) was based on a recently described flow cytometric method (O'Gorman 2002). Briefly, to measure phagocytic capacity 10 µL isothiocyanate-labeled *E. coli* and 100 µL whole blood were mixed and incubated at 37 °C for 10 min. Reaction was stopped by adding 100 µL ice-cold quenching solution (10 % Trypan in PBS). After washing, whole blood cells were fixed with 100 µL FACS-lysing solution (diluted 1:10, BD, Heidelberg, Germany). DNA was stained with 300 µL propidium iodide solution (33.3 mg/L, dissolved in PBS). A FACSCalibur flow cytometer (BD, Heidelberg) was used to measure the level of phagocytic activity and intensity in neutrophils and monocytes.

Statistical analyses

Results are reported as means ± standard error of the mean (SE). Changes between the baseline (0 h) and the following time-points among treatment groups was tested for significance by repeated-measures ANOVA with Fischer's test for comparison of individual means when appropriate. All statistical calculations were performed with the StatView program (SAS Institute 1998, Cary, NC, USA).

Results

The anthropometric data of the study subjects are summarized in Table 1. All participants tolerated the intervention with the 4 beverages very well and completed the study. Maximum blood ethanol concentrations in subjects consuming RW were 15.9 mM and 15.0 mM in subjects consuming ETOH.

The capacity of activated PBMC to produce the cytokines TNFα, IL-2, and IL-4 did not differ at baseline between subjects in the 4 groups. During the 24 h period following the consumption of the beverages, no specific effects were observed (Fig. 1). For TNFα and for IL-4, however, with all beverages a significant time-effect was seen ($p < 0.0001$ and $p = 0.029$, respectively). Phagocytic activity and intensity of neutrophils and monocytes ex vivo were not different at baseline and were not significantly affected by the consumption of the different beverages (Table 2). A significant time effect ($p < 0.0001$) was observed for phagocytic activity of neutrophils independent of the type of beverage consumed. Lymphocyte responsiveness to mitogen activation was also not affected by the beverages during the 24 h period (Table 3). The lytic activity of NK cells also showed for all beverages a significant time-effect ($p < 0.0001$), while there were no effects of the treatment on NK cell function (Table 3; data are only shown for the effector:target ratio of 25:1).

Discussion

Regular consumption of moderate amounts of wine is associated with a reduced risk of cardiovascular disease (Renaud et al. 1998, Rimm et al. 1999, Gaziano et al. 1999, Gronbæk et al. 2000) and of common cold (Takkouche et al. 2002). In the present study the immunomodulatory

Table 1 Anthropometric data of the study subjects (n = 6, mean ± SE)

Age (yr)	31 ± 1.6
Height (cm)	182 ± 2.9
Body mass (kg)	78 ± 4.9
BMI (kg/m^2)	24 ± 1.6

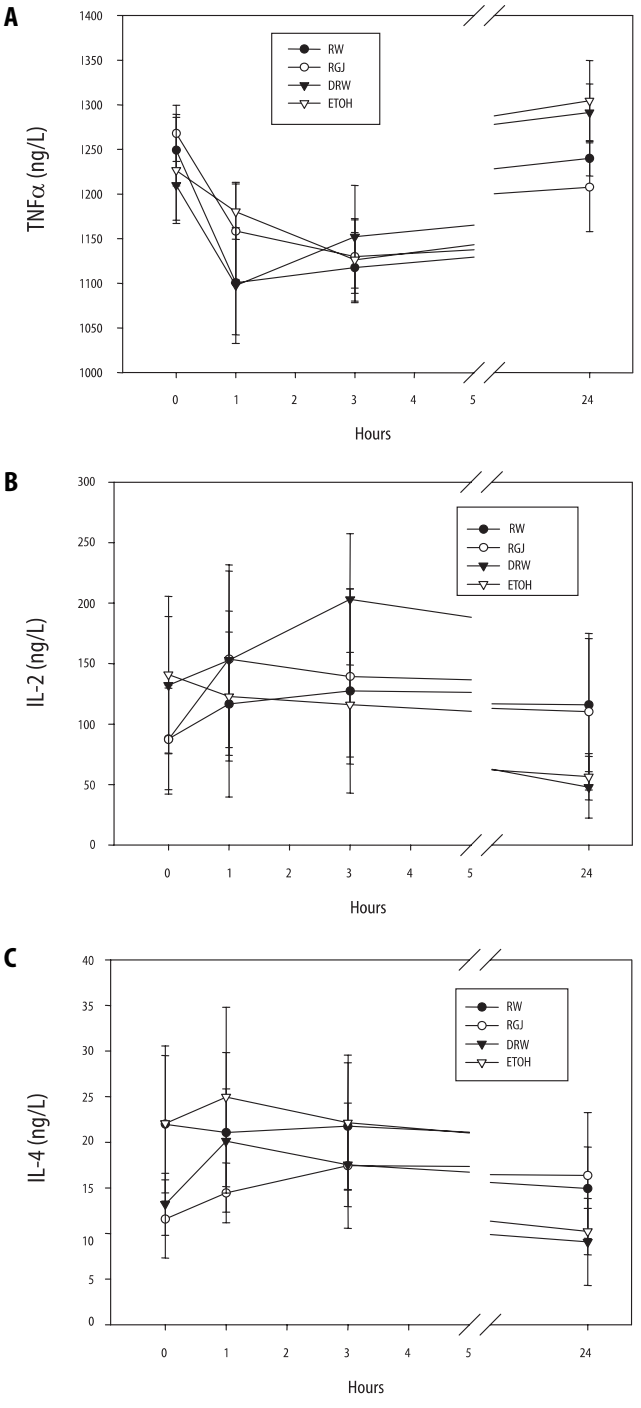

Fig. 1 Production of TNFα, IL-2, and IL-4 by activated peripheral blood mononuclear cells isolated from subjects consuming a single dose of 500 mL red wine (RW), dealcoholized red wine (DRW), red grape juice (RGJ) or a 12 % ethanol control beverage (ETOH) at time point 0, and 1, 3, and 24 h post intakes (n = 6, mean ± SE). **A** TNFα, **B** IL-2, **C** IL-4. A significant time-effect for TNFα and IL-4 was observed by ANOVA independent of the type of beverage consumed (TNFα, $p < 0.0001$; IL-4, $p = 0.03$)

Table 2 Phagocytic activity (percentage of phagocytic-active cells) and phagocytic intensity (mean fluorescence per phagocyte) of peripheral blood mononuclear cells isolated from subjects consuming a single dose of 500 mL red wine (RW), dealcoholized red wine (DRW), red grape juice (RGJ) or a 12 % ethanol control beverage (ETOH) at time point 0, and 1, 3, and 24 h post intakes (n = 6, mean ± SE)

	Time (h)			
	0	1	3	24
Phagocytic activity – neutrophils (%)[1]				
RW	40±4	41±4	54±5	53±5
DRW	35±5	37±6	42±4	47±3
RGJ	42±3	41±3	49±4	50±3
ETOH	41±3	41±4	49±5	50±2
Phagocytic intensity – neutrophils (mean fluorescence per neutrophil)				
RW	64±5	66±4	67±6	66±4
DRW	70±7	76±8	71±5	70±4
RGJ	63±3	66±6	67±5	67±2
ETOH	73±6	72±5	70±5	75±5
Phagocytic activity – monocytes (%)				
RW	26±3	27±3	27±4	28±3
DRW	26±6	26±6	26±5	24±3
RGJ	24±5	24±4	25±5	29±2
ETOH	23±3	20±3	22±4	25±4
Phagocytic intensity – monocytes (mean fluorescence per monocyte)				
RW	64±4	65±6	65±7	67±4
DRW	66±4	67±5	71±5	72±4
RGJ	65±6	70±6	63±4	63±4
ETOH	69±3	67±5	65±4	76±4

[1] A significant time-effect was observed by ANOVA independent of the type of beverage consumed ($p < 0.0001$)

Table 3 Proliferation and NK cell activity of peripheral blood mononuclear cells isolated from subjects consuming a single dose of 500 mL red wine (RW), dealcoholized red wine (DRW), red grape juice (RGJ) or a 12 % ethanol control beverage (ETOH) at time point 0, and 1, 3, and 24 h post intakes (n = 6, mean ± SE)

	Time (h)			
	0	1	3	24
Lymphocyte proliferation (A_{450}–A_{650})				
RW	0.96±0.09	0.90±0.09	1.12±0.11	0.91±0.23
DRW	1.03±0.14	0.86±0.13	0.91±0.15	0.90±0.17
RGJ	0.78±0.22	0.73±0.18	0.95±0.21	1.06±0.15
ETOH	1.07±0.13	1.04±0.07	1.17±0.16	1.09±0.10
NK cell activity (% lysed K562 target cells)[1, 2]				
RW	61.5±2.3	55.9±2.5	56.8±3.6	61.1±2.0
DRW	62.7±3.4	46.8±7.5	50.4±4.5	63.1±3.2
RGJ	64.3±4.0	55.5±9.0	56.0±7.0	59.0±4.8
ETOH	65.5±4.6	58.4±5.6	63.3±6.2	61.4±2.0

[1] Effector:target ratio 25:1
[2] A significant time-effect was observed by ANOVA independent of the type of beverage consumed ($p < 0.0001$)

effect of a single dose of RW or alcohol was studied. The rationale for such an approach was that we were interested in investigating polyphenol bioavailability from RW (Bub et al. 2001) as well as immunomodulatory effects of ethanol and polyphenols within the same study group during a 24 h period. This allowed us to relate changes in plasma polyphenol concentrations to

potential changes in immune parameters. The volume of 500 mL RW provided the study subjects with a reasonable amount of polyphenols without exposing them to adverse ethanol concentrations.

The major polyphenol in the beverages RW, DRW, and RGJ was the anthocyanin malvidin-3-glucoside, which contributed 80 % of the total anthocyanins in RW and DRW and 69 % in RGJ (Bub et al. 2001). The maximum malvidin-3-glucoside plasma concentrations in our study subjects were 1–3 nM, which were reached within 120 min following beverage consumption (Bub et al. 2001). This indicates that anthocyanins were absorbed, but only low plasma concentrations were achieved, possibly too low to induce immunological effects.

In order to study whether neutrophils, monocytes and lymphocytes were specifically affected by the different beverages, cell-specific functional assays were applied. Neutrophil functions such as chemotaxis and production of reactive oxygen species are known to be affected by ethanol and polyphenols (Patel et al. 1996, Lu et al. 2001). Acute ethanol exposure in humans (5 glasses of wine or more) induced neutrophil apoptosis (Singhal et al. 1999), which is expected to impair neutrophil functions. In the present study, however, no significant effects on neutrophil phagocytosis were observed with the different beverages. A significant time-effect with phagocytic activity was measured, resulting in increased activity 3 h and 24 h after the consumption of the 4 beverages. Currently, there is no explanation for this observation other than a systemic experimental effect.

No differences in phagocytic activity and intensity of monocytes were observed between the 4 beverages suggesting that neither ethanol nor red grape-associated polyphenols were effective. *In vitro*, ethanol exposure has been shown to attenuate the phagocytic activity of human monocytes via the Fc-receptor dose-dependently (Morland and Morland 1984). While concentrations comparable to those measured in the blood of our study subjects (12 or 22 mM) had no effect *in vitro* confirming our *in vivo* observations, higher *in vitro* ethanol concentrations significantly suppressed phagocytic activity (Morland and Morland 1984).

As a monocyte-derived cytokine we measured the production of TNFα by LPS-activated monocytes. No significant treatment effects were measured; however, a significant time-effect was observed. Such a circadian rhythm for TNFα has already been reported (Petrovsky et al. 1998). In another study with acute alcohol exposure (a single intake of 80 g ethanol with wine or beer) no changes in plasma concentrations of monocyte-derived cytokines such as TNFα and IL-1α/-β were measured (Mohadjer et al. 1995). *In vitro*, only high doses (= 25 mM) as well as pharmacological doses of ethanol (> 40 mM) significantly decreased TNFα production by human monocytes (Verma et al. 1993, Szabo et al. 1996, Arbabi et al. 1999). Blood ethanol concentrations in subjects of the present study may have been too low to affect LPS-induced TNFα production.

Lymphocyte-derived cytokine production (IL-2, IL-4) was likewise not modulated by ethanol or polyphenol intake. For IL-4, a TH2-lymphocyte-specific cytokine, no differences in serum levels between controls and alcoholic patients were observed in another study (Laso et al. 1998) supporting our observations that acute ethanol consumption has no effect on TH2-lymphocytes. However, binge drinking of up to 3.1 liters of beer significantly reduced IL-2 production, a TH1-specific cytokine (Bagasra et al. 1989). In an animal study, a long-term intake of high ethanol doses significantly increased IL-2 production independent of dietary composition (Watzl et al. 1993). This suggests that, in contrast to TH2-lymphocytes, ethanol modulates TH1-lymphocytes depending on dose and intake period. Lymphocyte proliferative responsiveness to mitogen activation was also not influenced by the different beverages. This is in agreement with an earlier human study, which did not observe acute effects of ethanol after alcohol consumption (Mohadjer et al. 1995). *In vitro*, various flavonoids at a concentration of 1 μM showed no effect on ConA-activated lymphocyte proliferation of mouse splenocytes (Namgoong et al. 1993).

Lytic activity of NK cells was not affected by ETOH or by RW. Another study with acute ethanol consumption (blood ethanol levels 8.7 mM and 19.3 mM) also found no change in NK cell lytic activity (Ochshorn-Adelson et al. 1994). In contrast, chronic alcoholics without liver disease were shown to have a significantly increased number of NK cells and a parallel increase in NK cell lytic activity (Laso et al. 1997). Recently, interesting data from an animal study were published showing that long-term ethanol consumption (14 % of the total daily energy intake) reduced NK cell numbers, which was not seen in mice exposed to a similar dose of ethanol taken up from red wine (Percival and Sims 2000). In this animal model red wine constituents may overcome the detrimental effects of high ethanol concentrations on NK cells. The circadian rhythm in NK cell lytic activity observed in the present study has also been reported by others (Haus 1996). Some studies have investigated the effect of wine flavonoids on NK cell function. While quercetin *in vitro* (1 mM) significantly decreased lytic activity of NK cells (no effects at lower concentrations) (Exon et al. 1998), in animal models quercetin (100 mg/kg b. w.) and catechin (125–500 mg/kg b. w.) increased lytic activity of NK cells (Ikeda et al. 1984, Exon et al. 1998). The doses used in *in vitro* studies and those obtained in the animal studies were much higher than the plasma polyphenol concentrations in the present study.

In conclusion, the results of the present study clearly

show that in healthy men acute intake of different alcoholic beverages at a volume of 500 ml has no significant immunomodulatory effects in the 24 h following beverage consumption. In contrast to the results from several *in vitro* studies (Middleton et al. 2000), a moderate polyphenol intake with RGJ and DRW did not result in immunosuppression. Whether long-term exposure to moderate amounts of alcohol and RW significantly affects immunity in man has to be investigated in future studies.

Acknowledgments We gratefully acknowledge the competent technical assistance of the IEP technicians and are indebted to all study participants for contributing continuously to this study. The study was supported by a special grant from the Federal Ministry of Consumer Protection, Food, and Agriculture, Germany.

References

1. Arbabi S, Garcia I, Bauer GJ, Maier RV (1999) Alcohol (ethanol) inhibits IL-8 and TNF: role of the p38 pathway. J Immunol 162:7441–7445
2. Bagasra O, Kajdacsy-Balla A, Lischner HW (1989) Effects of alcohol ingestion on in vitro susceptibility of peripheral blood mononuclear cells to infection with HIV and of selected T-cell functions. Alcohol Clin Exp Res 13:636–643
3. Bofetta P, Garfinkel L (1990) Alcohol drinking and mortality among men enrolled in an American Cancer Society prospective study. Epidemiology 1: 342–348
4. Bub A, Watzl B, Heeb D, Rechkemmer G, Briviba K (2001) Malvidin-3-glucoside bioavailability in humans after ingestion of red wine, dealcoholized red wine and red grape juice. Eur J Nutr 40: 113–120
5. Casalini C, Lodovici M, Briani C, Paganelli G, Remy S, Cheynier V, Dolara P (1999) Effect of complex polyphenols and tannins from red wine (WCPT) on chemically induced oxidative DNA damage in the rat. Eur J Nutr 38: 190–195
6. Cohen S, Tyrell DA, Russell MA, Jarvis MJ, Smith AP (1993) Smoking, alcohol consumption, and susceptibility to the common cold. Am J Public Health 83: 1277–1283
7. Exon JH, Magnuson BA, South EH, Hendrix K (1998) Dietary quercetin, immune functions and colonic carcinogenesis in rats. Immunopharmacol Immunotoxicol 20:173–190
8. Gaziano JM, Hennekens CH, Godfried SL, Sesso HD, Glynn RJ, Breslow JL, Buring JE (1999) Type of alcoholic beverage and risk of myocardial infarction. Am J Cardiol 83:52–57
9. Gronbœk M, Becker U, Johansen D, Gottschau A, Schnohr P, Hein HO, Jensen G, Sorensen TIA (2000) Type of alcohol consumed and mortality from all causes, coronary heart disease, and cancer. Ann Intern Med 133:411–419
10. Haus E (1996) Biologic rhythms in hematology. Path Biol 44:618–630
11. Ikeda Y, Kawakami K, Sato I, Tajima S, Ito K, Nose S (1984) Effect of cianidanol (KB-53) on the activity of mouse cytotoxic T-lymphocytes. J Pharmacobiodyn 7:15–20
12. Laso FJ, Madruga JI, Giron JA, Lopez A, Ciudad J, San Miguel JF, Alvarez-Mon M, Orfao A (1997) Decreased natural killer cell cytotoxic activity in chronic alcoholism is associated with alcohol liver disease but not active ethanol consumption. Hepatology 26:1096–1100
13. Laso FJ, Iglesias MC, Lopez A, Ciudad J, San Miguel JF, Orfao A (1998) Increased interleukin-12 serum levels in chronic alcoholism. J Hepatol 28:771–777
14. Lodovici M, Guglielmi F, Casalini C, Meoni M, Cheynier V, Dolara P (2001) Antioxidant and radical scavenging properties in vitro of polyphenolic extracts from red wine. Eur J Nutr 40:74–77
15. Lu HW, Sugahara K, Sagara Y, Masuoka N, Asaka Y, Manabe M, Kodama H (2001) Effect of three flavonoids, 5,7,3',4'-tetrahydroxy-3-methoxy flavone, luteolin, and quercetin, on the stimulus-induced superoxide generation and tyrosyl phosphorylation of proteins in human neutrophil. Arch Biochem Biophys 393:73–77
16. Middleton E, Kandaswami C, Theoharides TC (2000) The effects of plant flavonoids on mammalian cells: implications for inflammation, heart disease and cancer. Pharmacol Rev 52:673–751
17. Mohadjer C, Daniel V, Althof F, Maier H (1995) Immunological status of healthy subjects after a single intake of alcohol. Dtsch Med Wschr 120:1577–1581
18. Morland B, Morland J (1984) Reduced Fc-receptor function in human monocytes exposed to ethanol in vitro. Alcohol Alcohol 19:211–217
19. Namgoong SY, Son KH, Chang HW, Kang SS, Kim HP (1993) Effects of naturally-occurring flavonoids on mitogen-induced lymphocyte proliferation and mixed lymphocyte culture. Life Sci 54:313–320
20. Nelson S, Kolls JK (2002) Alcohol, host defence and society. Nature Rev Immunol 2:205–209
21. Ochshorn-Adelson M, Bodner G, Toraker P, Albeck H, Ho A, Kreek MJ (1994) Effects of ethanol on human natural killer cell activity: in vitro and acute, low-dose in vivo studies. Alcohol Clin Exp Res 18:1361–1367
22. O'Gorman MR (2002) Evaluation of phagocytic cell function. In: Rose NR, Hamilton, RG, Detrick, B (eds) Manual of Clinical Laboratory Immunology, 6th edn. ASM Press, Washington, pp 265–273
23. Patel M, Keshavarzian A, Kottapalli V, Badie B, Winship D, Fields JZ (1996) Human neutrophil functions are inhibited in vitro by clinically relevant ethanol concentrations. Alcohol Clin Exp Res 20:275–283
24. Percival SS, Sims CA (2000) Wine modifies the effects of alcohol on immune cells of mice. J Nutr 130:1091–1094
25. Petrovsky N, McNair P, Harrison LC (1998) Diurnal rhythms of pro-inflammatory cytokines: regulation by plasma cortisol and therapeutic implications. Cytokine 10:307–312
26. Renaud SC, Guéguen R, Schenker J, d'Houtaud A (1998) Alcohol and mortality in middle-aged men from eastern France. Epidemiology 9:184–188
27. Rimm EB, Williams P, Fosher K, Criqui M, Stampfer MJ (1999) Moderate alcohol intake and lower risk of coronary heart disease: meta-analysis of effects on lipids and haemostatic factors. BMJ 319:1523–1528
28. Singhal PC, Reddy K, Ding G, Kapasi A, Franki N, Ranjan R, Nwakoby IE, Gibbson N (1999) Ethanol-induced macrophage apoptosis: the role of TGF-β. J Immunol 162:3031–3036
29. Stoltz DA, Zhang P, Nelson S, Bohm RP, Murphey-Corb M, Bagby GJ (1999) Ethanol suppression of the functional state of polymorphonuclear leukocytes obtained from uninfected and simian immunodeficiency virus infected rhesus macaques. Alcohol Clin Exp Res 23: 878–894
30. Szabo G (1999) Consequences of alcohol consumption on host defence. Alcohol Alcohol 34:830–841

31. Szabo G, Mandrekar P, Girouard L, Catalano D (1996) Regulation of human monocyte functions by acute ethanol treatment: decreased tumor necrosis factor-alpha, interleukin-1beta and elevated interleukin-10, and transforming growth factor-beta production. Alcohol Clin Exp Res 20:900–907
32. Takkouche B, Regueira-Méndez C, Garcia-Closas R, Figueiras A, Gestal-Otero JJ, Hernán MA (2002) Intake of wine, beer, and spirits and the risk of clinical common cold. Am J Epidemiol 155: 853–858
33. Teissedre PL, Landrault N (2000) Wine phenolics: contribution of dietary intake and bioavailability. Food Res Int 33:461–467
34. Verma BK, Fogarasi M, Szabo G (1993) Down-regulation of TNFα activity by acute ethanol treatment in human peripheral blood monocytes. J Clin Immunol 13:8–22
35. Wang H, Cao G, Prior RL (1997) Oxygen radical absorbing capacity of anthocyanins. J Agric Food Chem 45:304–309
36. Watzl B, Watson RR (1992) Role of alcohol abuse in nutritional immunosuppression. J Nutr 122:733–737
37. Watzl B, Watson RR (1993) Alcohol and cytokine secretion. In: Yirmiya R, Taylor AN (eds) Alcohol, Immunity, and Cancer. CRC Press, Boca Raton, pp 87–101
38. Watzl B, Lopez M, Shabazian M, Chen G, Colombo LL, Huang DS, Way D, Watson RR (1993) Diet and ethanol modulate immune response in young C57BL/6 mice. Alcohol Clin Exp Res 17:623–630
39. Watzl B, Bub A, Blockhaus M, Herbert BM, Lührmann PM, Neuhäuser-Berthold M, Rechkemmer G (2000) Prolonged tomato juice consumption has no effect on cell-mediated immunity of well-nourished elderly men and women. J Nutr 30:1719–1723