

1 **Supplementary Information for**
2 **Very-light alcohol consumption suppresses breast tumor**
3 **progression in mouse model**

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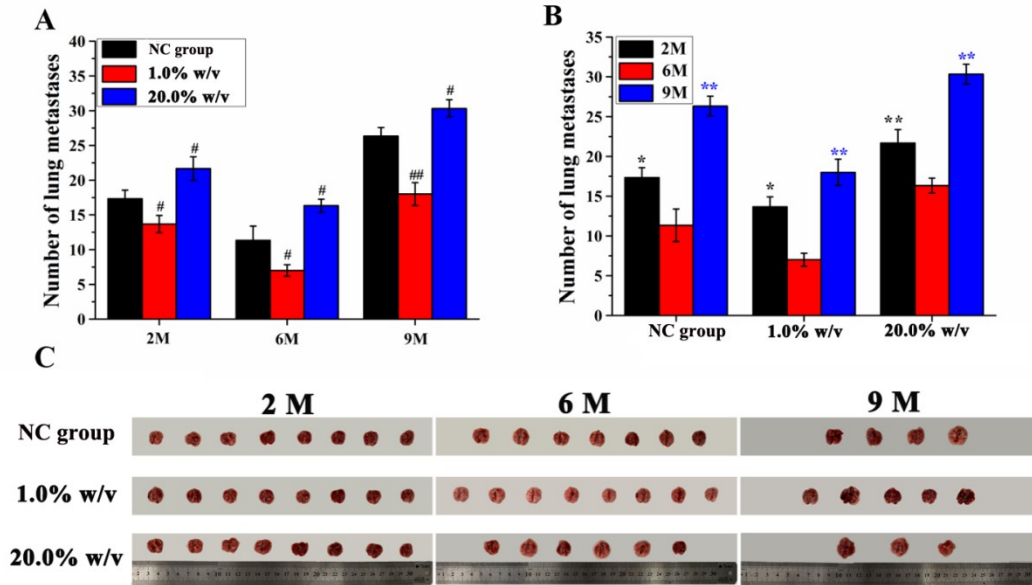
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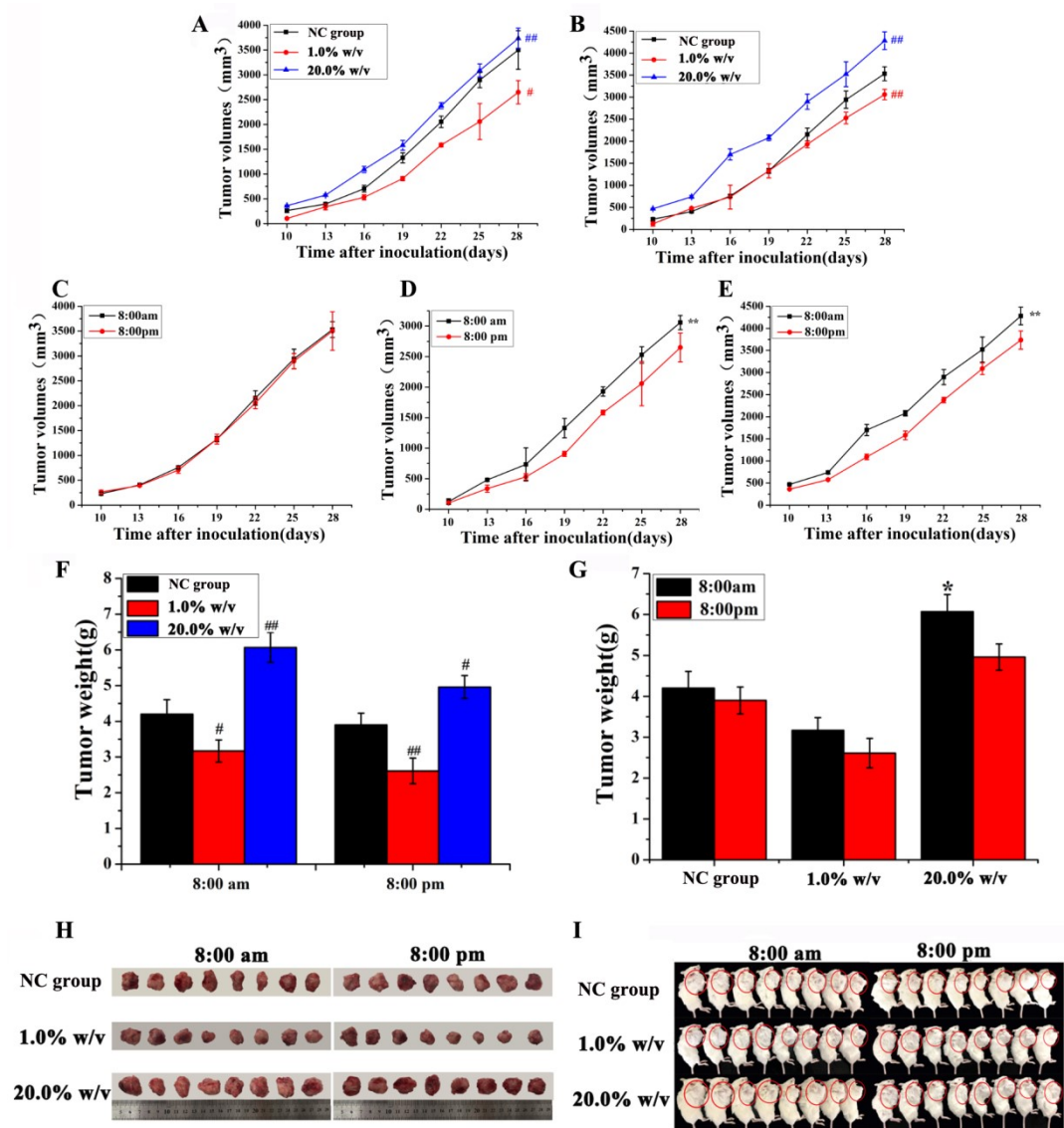
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Supplementary Fig. S1



Supplementary Fig. S1 The effect of age on breast tumor metastasis at the dosage of 1.0% w/v group and 20.0% w/v group. (A) Number of lung metastases of different age groups at the dosage of 1.0% w/v and 20.0% w/v. $\#p < 0.05$ and $\#\#p < 0.01$ compared with control group. (B) Number of lung metastases of different alcohol consumption dosages groups at the age of 2-month, 6-month and 9-month. $*p < 0.05$ and $**p < 0.01$ compared with 6-month adult group. (C) Images of excised lungs. N value of each group is the number of mice left on the 35th day.

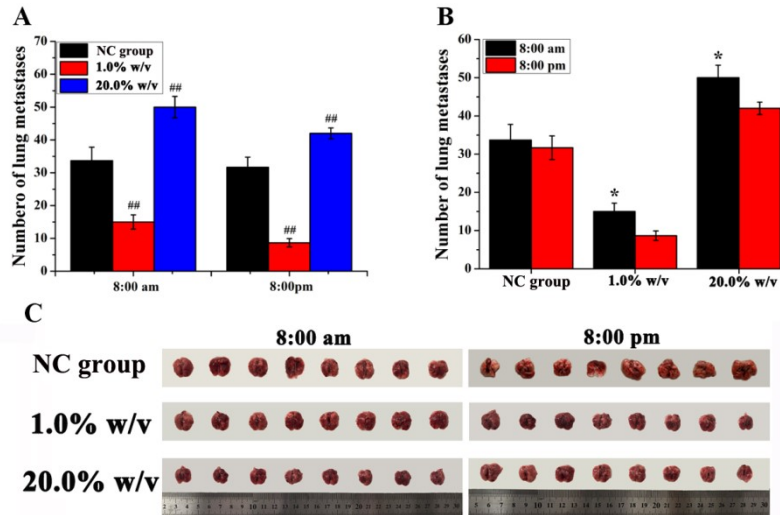
Supplementary Fig. S2



Supplementary Fig. S2 The effect of time of drinking on breast primary tumors growth at the dosage of 1.0% w/v group and 20.0% w/v group. Breast primary tumor volumes in (A) 8:00 a.m. group and (B) 8:00 p.m. group with different alcohol consumption dosages. Values were presented as mean \pm SD, n = 8. #p < 0.05 and ##p < 0.01 compared with control group. Breast primary tumor volumes in (D) control group, (E) 1.0% w/v group and (F) 20.0% w/v group with different time of drinking. Values were presented as mean \pm SD, n = 8. *p < 0.05 and **p < 0.01 compared with

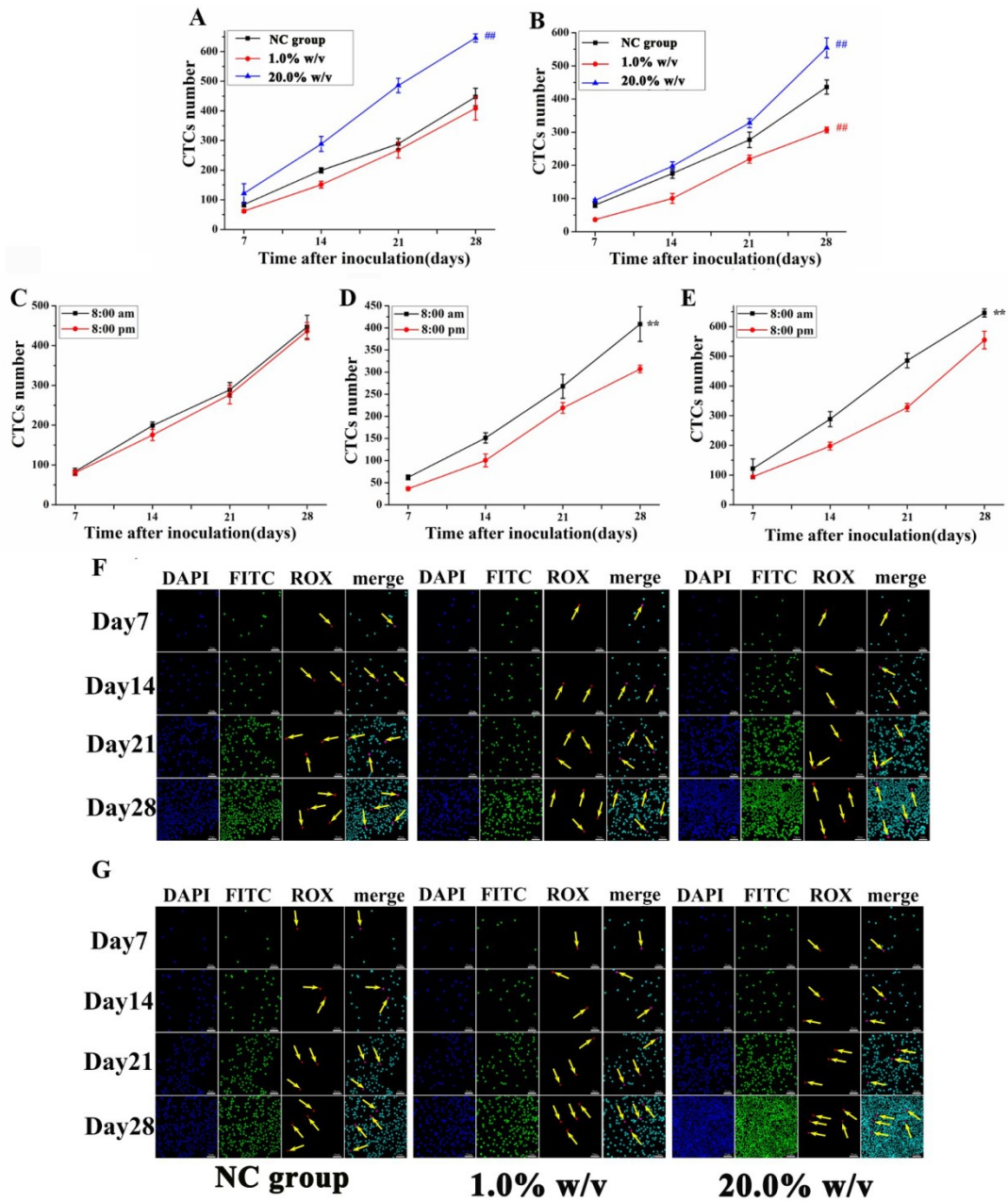
8:00 a.m. group. (G) Tumor weight of different time of drinking groups at the dosage of 1.0% w/v and 20.0% w/v recorded on day 35 (n=8), #p < 0.05 and ##p < 0.01 compared with control group. (H) Tumor weight of different alcohol consumption dosages groups with time of drinking of 8:00 a.m. and 8:00 p.m. recorded on day 35 (n=8), *p < 0.05 and **p < 0.01 compared with 8:00 a.m. group. (I) Images of excised tumors on day 35, n=8. (J) Images of *in vivo* tumors, n=8.

Supplementary Fig. S3



Supplementary Fig. S3. The effect of time of drinking on breast tumor metastasis at the dosage of 1.0% w/v group and 20.0% w/v group. (A) Number of lung metastases of different time of drinking groups at the dosage of 1.0% w/v and 20.0% w/v, n=8. #p < 0.05 and ##p < 0.01 compared with control group. (B) Number of lung metastases of different alcohol consumption dosages groups with time of drinking of 8:00 a.m. and 8:00 p.m., n=8. *p < 0.05 and **p < 0.01 compared with 8:00 a.m. group. (C) Images of excised lungs, n=8.

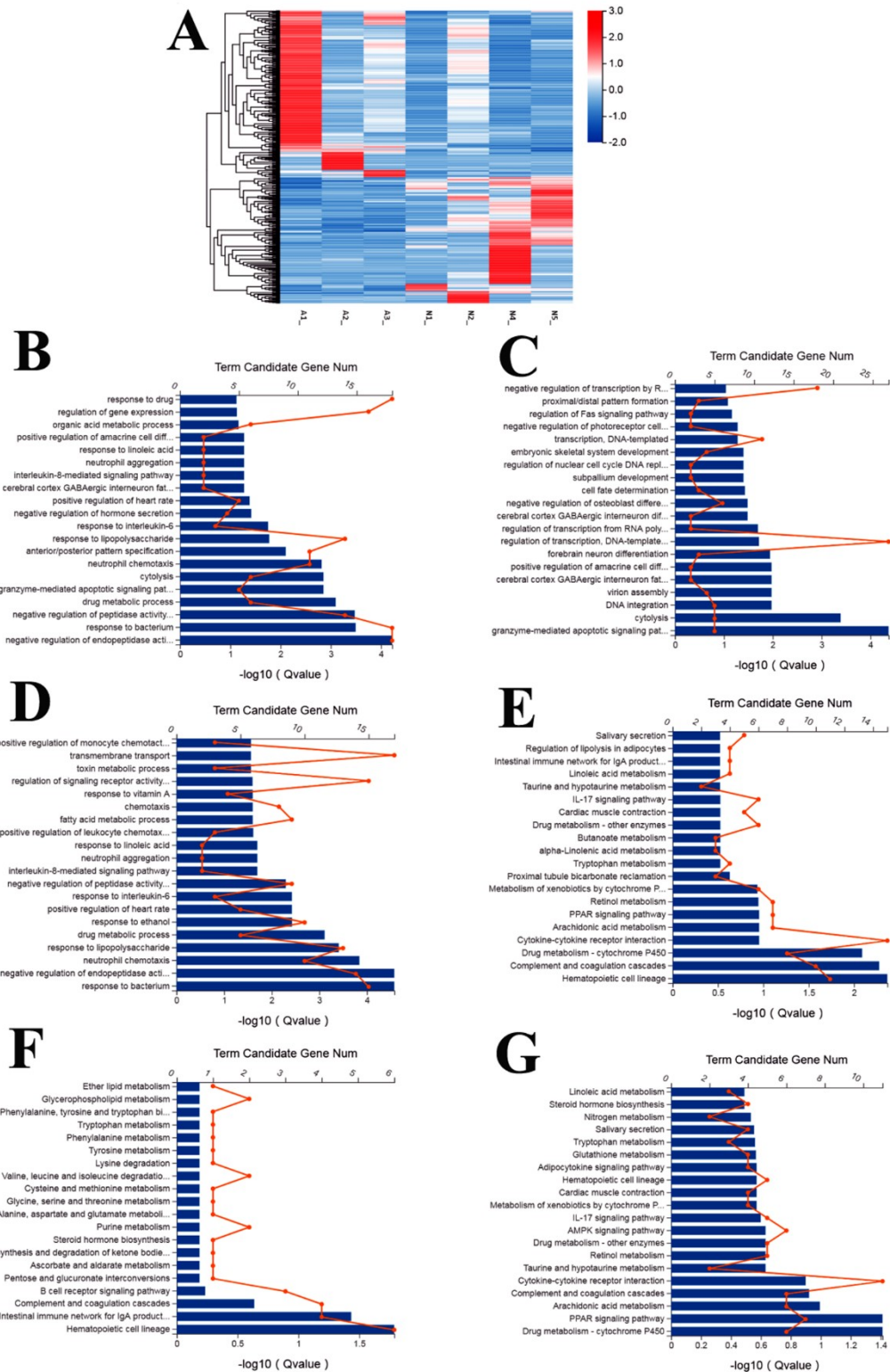
Supplementary Fig. S4



Supplementary Fig. S4. The effect of time of drinking on numbers of CTCs at the dosage of 1.0% w/v group and 20.0% w/v group. CTC numbers in (A) 8:00 a.m. and (B) 8:00 p.m. group with different alcohol consumption dosages. Values were presented as mean \pm SD, n = 8. #p < 0.05 and ##p < 0.01 compared with control group. CTC numbers in (C) control group, (D) 1.0% w/v group and (E) 20.0% w/v

group with different time of drinking. Values were presented as mean \pm SD, n = 8. *p < 0.05 and **p < 0.01 compared with 8:00 a.m. group. Fluorescence images of *in vivo* captured CTCs during cancer progression at the dosage of 1.0% w/v and 20.0% w/v of (F) 8:00 a.m. group and (G) 8:00 p.m. group.

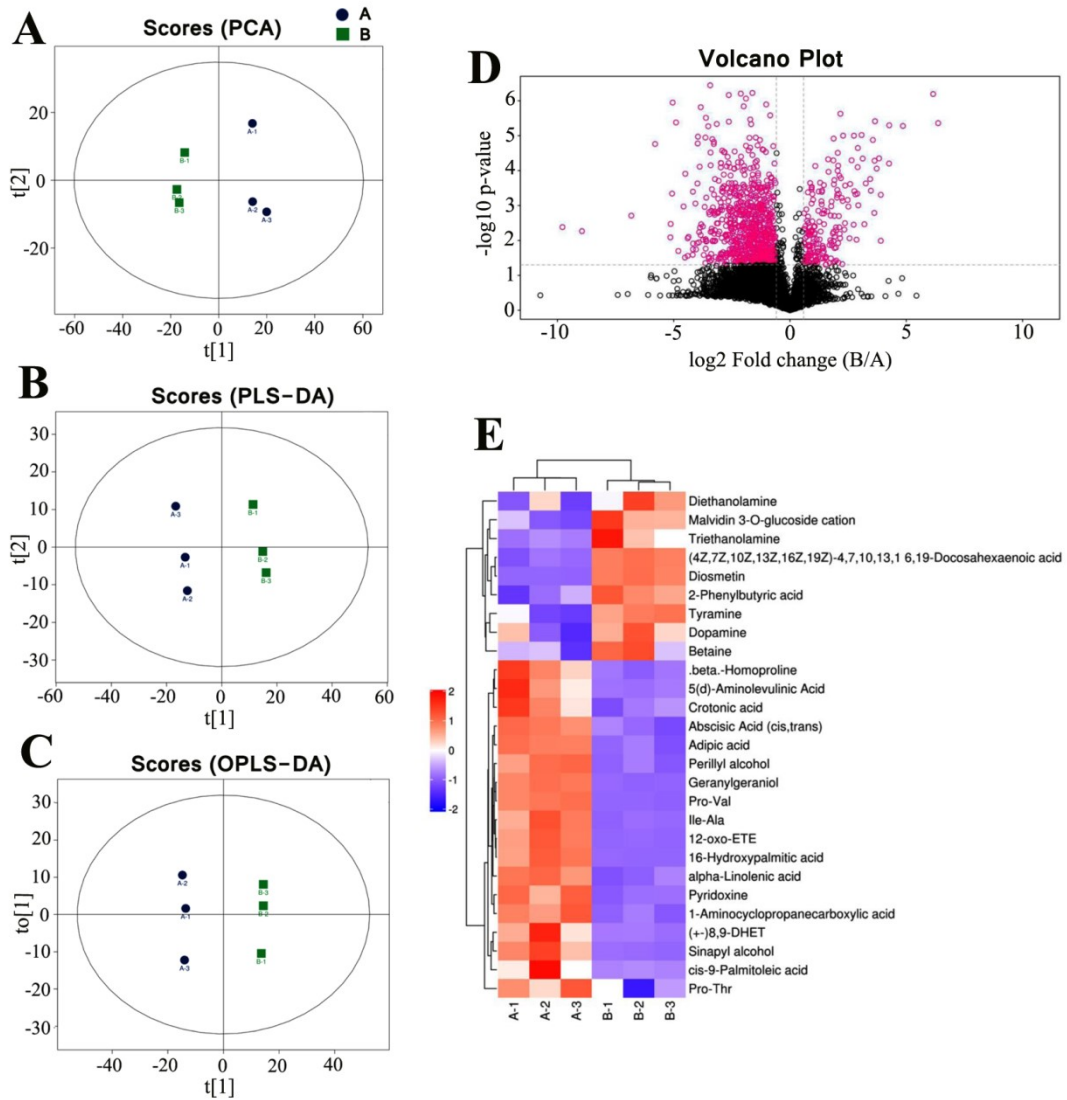
Supplementary Fig. S5



Supplementary Fig. S5. RNA-seq analysis in metastatic carcinoma from NC group and 1.0% w/v ($\sim 4.38 \times 10^{-2}$ g/kg/day, 2-month old mice, fed at 8:00 p.m.) group. (A)

Heatmap of differentially expressed genes (n = 3, biological replicates), (B) GO analysis of all differentially expressed genes, (C) GO analysis of downregulated expressed genes in 1.0% w/v group versus NC group, (D) GO analysis of upregulated expressed genes in 1.0% w/v group versus NC group, (E) KEGG analysis of all differentially expressed genes, (F) KEGG analysis of downregulated expressed genes in 1.0% w/v group versus NC group, (G) KEGG analysis of upregulated expressed genes in 1.0% w/v group versus NC group.

Supplementary Fig. S6



Supplementary Fig. S6. Components comparison analysis of red wine (brand 1) and red wine (brand 2). (A) PCA analysis, (B) PLS-DA analysis, (C) OPLS-DA analysis, (D) Volcano plot for red wine (brand 1) group versus red wine (brand 2) group, (E) Heatmap for the 27 difference compounds from red wine (brand 1) group and red wine (brand 2) group.